```
$%^STN;HighlightOn= ***;HighlightOff=*** ;
Trying 3106016892...Open

Welcome to STN International!  Enter x:x
LOGINID:ssspta1633cxq
PASSWORD:
TERMINAL (ENTER 1, 2, 3, OR ?):2


********** Welcome to STN International **********

NEWS 1       Web Page URLs for STN Seminar Schedule - N. America
NEWS 2 Dec 17 The CA Lexicon available in the CAPLUS and CA files
NEWS 3 Feb 06 Engineering Information Encompass files have new names
NEWS 4 Feb 16 TOXLINE no longer being updated
NEWS 5 Apr 23 Search Derwent WPINDEX by chemical structure
NEWS 6 Apr 23 PRE-1967 REFERENCES NOW SEARCHABLE IN CAPLUS
AND CA
NEWS 7 May 07 DGENE Reload
NEWS 8 Jun 20 Published patent applications (A1) are now in USPATFULL
NEWS 9 JUL 13 New SDI alert frequency now available in Derwent's
          DWPI and DPCI
NEWS 10 Aug 23 In-process records and more frequent updates now in
          MEDLINE
NEWS 11 Aug 23 PAGE IMAGES FOR 1947-1966 RECORDS IN CAPLUS AND
CA
NEWS 12 Aug 23 Adis Newsletters (ADISNEWS) now available on STN
NEWS 13 Sep 17 IMSworld Pharmaceutical Company Directory name change
          to PHARMASEARCH

NEWS EXPRESS  August 15 CURRENT WINDOWS VERSION IS V6.0c,
          CURRENT MACINTOSH VERSION IS V6.0 (ENG) AND V6.0J (JP),
          AND CURRENT DISCOVER FILE IS DATED 07 AUGUST 2001
NEWS HOURS   STN Operating Hours Plus Help Desk Availability
NEWS INTER   General Internet Information
NEWS LOGIN   Welcome Banner and News Items
NEWS PHONE   Direct Dial and Telecommunication Network Access to STN
NEWS WWW     CAS World Wide Web Site (general information)

Enter NEWS followed by the item number or name to see news on that
specific topic.

  All use of STN is subject to the provisions of the STN Customer
  agreement.  Please note that this agreement limits use to scientific
  research.  Use for software development or design or implementation
  of commercial gateways or other similar uses is prohibited and may
  result in loss of user privileges and other penalties.

  ********** STN Columbus **********


  FILE 'HOME' ENTERED AT 09:34:11 ON 18 SEP 2001

  => file biosis embase caplus
  COST IN U.S. DOLLARS                 SINCE FILE     TOTAL
                                 ENTRY   SESSION
  FULL ESTIMATED COST                      0.15      0.15

  FILE 'BIOSIS' ENTERED AT 09:34:29 ON 18 SEP 2001
  COPYRIGHT (C) 2001 BIOSIS(R)

  FILE 'EMBASE' ENTERED AT 09:34:29 ON 18 SEP 2001
  COPYRIGHT (C) 2001 Elsevier Science B.V.  All rights reserved.

  FILE 'CAPLUS' ENTERED AT 09:34:29 ON 18 SEP 2001
  USE IS SUBJECT TO THE TERMS OF YOUR STN CUSTOMER AGREEMENT.
  PLEASE SEE "HELP USAGETERMS" FOR DETAILS.
  COPYRIGHT (C) 2001 AMERICAN CHEMICAL SOCIETY (ACS)

  => s LDLR354
  L1          0 LDLR354

  => s LDLR 354
  L2          0 LDLR 354

  => s LDLR
  L3       1661 LDLR

  => s KDEL
  L4        879 KDEL

  => s KEEL
  L5        910 KEEL

  => s HDEL
  L6        263 HDEL

  => s DDEL
  L7         78 DDEL

  => s QDEL
  L8          9 QDEL

  => s ADEL
  L9         59 ADEL

  => s SDEL
  L10        16 SDEL

  => d his

     (FILE 'HOME' ENTERED AT 09:34:11 ON 18 SEP 2001)

     FILE 'BIOSIS, EMBASE, CAPLUS' ENTERED AT 09:34:29 ON 18 SEP 2001
  L1          0 S LDLR354
  L2          0 S LDLR 354
  L3       1661 S LDLR
  L4        879 S KDEL
  L5        910 S KEEL
  L6        263 S HDEL
  L7         78 S DDEL
  L8          9 S QDEL
  L9         59 S ADEL
  L10        16 S SDEL

  => s fusion or chimeric or hybrid or heterologous
  L11    571052 FUSION OR CHIMERIC OR HYBRID OR HETEROLOGOUS

  => s l3 and l4 and l11
  L12         0 L3 AND L4 AND L11

  => s l3 and l4
  L13         0 L3 AND L4

  => s l3 and l5 and l11
  L14         0 L3 AND L5 AND L11

  => s l3 and l6 and l11
  L15         0 L3 AND L6 AND L11

  => s l3 and l7
  L16         0 L3 AND L7

  => s l3 and l8
  L17         0 L3 AND L8

  => s l3 and l9
  L18         0 L3 AND L9

  => s l3 and l10
  L19         0 L3 AND L10

  => d his

     (FILE 'HOME' ENTERED AT 09:34:11 ON 18 SEP 2001)

     FILE 'BIOSIS, EMBASE, CAPLUS' ENTERED AT 09:34:29 ON 18 SEP 2001
  L1          0 S LDLR354
  L2          0 S LDLR 354
  L3       1661 S LDLR
  L4        879 S KDEL
  L5        910 S KEEL
  L6        263 S HDEL
  L7         78 S DDEL
  L8          9 S QDEL
  L9         59 S ADEL
  L10        16 S SDEL
  L11    571052 S FUSION OR CHIMERIC OR HYBRID OR HETEROLOGOUS
  L12         0 S L3 AND L4 AND L11
  L13         0 S L3 AND L4
  L14         0 S L3 AND L5 AND L11
  L15         0 S L3 AND L6 AND L11
  L16         0 S L3 AND L7
  L17         0 S L3 AND L8
  L18         0 S L3 AND L9
  L19         0 S L3 AND L10

  => s l3 and l11
  L20        81 L3 AND L11

  => s l4 and l11
  L21       195 L4 AND L11

  => s l5 and l11
  L22        40 L5 AND L11

  => s l6 and l11
  L23        60 L6 AND L11

  => s l7 and l11
  L24         3 L7 AND L11

  => s l8 and l11
  L25         3 L8 AND L11
```

```
=> s l9 and l11
L26         5 L9 AND L11

=> s l10 and l11
L27         1 L10 AND L11

=> d his

     (FILE 'HOME' ENTERED AT 09:34:11 ON 18 SEP 2001)

     FILE 'BIOSIS, EMBASE, CAPLUS' ENTERED AT 09:34:29 ON 18 SEP 2001
L1          0 S LDLR354
L2          0 S LDLR 354
L3       1661 S LDLR
L4        879 S KDEL
L5        910 S KEEL
L6        263 S HDEL
L7         78 S DDEL
L8          9 S QDEL
L9         59 S ADEL
L10        16 S SDEL
L11    571052 S FUSION OR CHIMERIC OR HYBRID OR HETEROLOGOUS
L12         0 S L3 AND L4 AND L11
L13         0 S L3 AND L4
L14         0 S L3 AND L5 AND L11
L15         0 S L3 AND L6 AND L11
L16         0 S L3 AND L7
L17         0 S L3 AND L8
L18         0 S L3 AND L9
L19         0 S L3 AND L10
L20        81 S L3 AND L11
L21       195 S L4 AND L11
L22        40 S L5 AND L11
L23        60 S L6 AND L11
L24         3 S L7 AND L11
L25         3 S L8 AND L11
L26         5 S L9 AND L11
L27         1 S L10 AND L11

=> dup rem l20
PROCESSING COMPLETED FOR L20
L28        34 DUP REM L20 (47 DUPLICATES REMOVED)

=> d bib abs

L28  ANSWER 1 OF 34  BIOSIS  COPYRIGHT 2001 BIOSIS      DUPLICATE 1
AN   2001:354346  BIOSIS
DN   PREV200100354346
TI   Differential functions of members of the low density lipoprotein receptor
     family suggested by their distinct endocytosis rates.
AU   Li, Yonghe; Lu, Wenyan; Marzolo, Maria Paz; Bu, Guojun (1)
CS   (1) Dept. of Pediatrics, Washington University School of Medicine, 660
     South Euclid Ave., St. Louis, MO, 63110: bu@kids.wustl.edu USA
SO   Journal of Biological Chemistry, (May 25, 2001) Vol. 276, No. 21, pp.
     18000-18006. print.
     ISSN: 0021-9258.
DT   Article
LA   English
SL   English
AB   The low density lipoprotein receptor ( ***LDLR*** ) family is composed
     of a class of cell surface endocytic receptors that recognize
     extracellular ligands and internalize them for degradation by lysosomes.
     In addition to ***LDLR*** , mammalian members of this family include
     the ***LDLR*** -related protein (LRP), the very low density lipoprotein
     receptor (VLDLR), the alipoprotein E receptor-2 (apoER2), and megalin.
     Herein we have analyzed the endocytic functions of the cytoplasmic tails
     of these receptors using LRP minireceptors, its ***chimeric***
     receptor constructs, and full-length VLDLR and apoER2 stably expresseed in
     LRP-null Chinese hamster ovary cells. We find that the initial endocytosis
     rates mediated by different cytoplasmic tails are significantly different,
     with half-times of ligand internalization ranging from less than 30 s to
     more than 8 min. The tail of LRP mediates the highest rate of endocytosis,
     whereas those of the VLDLR and apoER2 exhibit least endocytosis function.
     Compared with the tail of LRP, the tails of the ***LDLR*** and megalin
     display significantly lower levels of endocytosis rates. Ligand
     degradation analyses strongly support differential endocytosis rates
     initiated by these receptors. Interestingly, apoER2, which has recently
     been shown to mediate intracellular signal transduction, exhibited the
     lowest level of ligand degradation efficiency. These results thus suggest
     that the endocytic functions of members of the ***LDLR*** family are
     distinct and that certain receptors in this family may play their main
     roles in areas other than receptor-mediated endocytosis.


=> s low density lipoprotein receptor or LDLR or (LDLR and 354)
L29      9513 LOW DENSITY LIPOPROTEIN RECEPTOR OR LDLR OR (LDLR
AND 354)

=> s l29 and l4
L30         3 L29 AND L4

=> dup rem l30
PROCESSING COMPLETED FOR L30
L31         2 DUP REM L30 (1 DUPLICATE REMOVED)

=> d bib abs 1-
YOU HAVE REQUESTED DATA FROM 2 ANSWERS - CONTINUE? Y/(N):y

L31  ANSWER 1 OF 2  BIOSIS  COPYRIGHT 2001 BIOSIS
AN   2001:113521  BIOSIS
DN   PREV200100113521
TI   Direct evidence that the ***low***  ***density***
     ***lipoprotein***  ***receptor***  regulates apolipoprotein B
     secretion in the secretory pathway.
AU   Gillian-Daniel, Donald L. (1); Bates, Paul W. (1); Tebon, Angie (1);
     Attie, Alan D. (1)
CS   (1) Univ of Wisconsin, Madison, WI USA
SO   Circulation, (October 31, 2000) Vol. 102, No. 18 Supplement, pp. II.148.
     print.
     Meeting Info.: Abstracts from Scientific Sessions 2000 New Orleans,
     Louisiana, USA November 12-15, 2000
     ISSN: 0009-7322.
DT   Conference
LA   English
SL   English

L31  ANSWER 2 OF 2  BIOSIS  COPYRIGHT 2001 BIOSIS      DUPLICATE 1
AN   1997:107420  BIOSIS
DN   PREV199799406623
TI   ERD2 proteins mediate ER retention of the HNEL signal of LRP's
     receptor-associated protein (RAP.
AU   Bu, Guojun (1); Rennke, Stephanie; Geuze, Hans J.
CS   (1) Dep. Pediatr., Washington Univ. Sch. Med., St. Louis, MO 63110 USA
SO   Journal of Cell Science, (1997) Vol. 110, No. 1, pp. 65-73.
     ISSN: 0021-9533.
DT   Article
LA   English
AB   The 39 kDa receptor-associated protein (RAP) is a receptor antagonist that
     interacts with several members of the low density lipoprotein (LDL)
     receptor gene family. Upon binding to these receptors, RAP inhibits all
     ligand interactions with the receptors. Our recent studies have
     demonstrated that RAP is an endoplasmic reticulum (ER) resident protein
     and an intracellular chaperone for the LDL receptor-related protein (LRP).
     The HNEL sequence at the carboxyl terminus of RAP represents a novel ER
     retention signal that shares homology with the well-characterized
     ***KDEL*** signal. In the present study, using immunoelectron microscopy
     we demonstrate that cells stably transfected with human growth hormone
     (GH) tagged with either ***KDEL*** (GH+ ***KDEL*** ) or HNEL
     (GH+HNEL) signals exhibit ER and cis-Golgi localization typical of
     ER-retained proteins. Overexpression of not only GH+HNEL but also GH+
     ***KDEL*** cDNA in transfected cells results in saturation of ER
     retention receptors and secretion of endogenous RAP indicating that the
     two signals interact with the same ER retention receptors. The role of RAP
     in the maturation of LRP is further supported by the observation that
     functional LRP is reduced about 60% as a result of decreased intracellular
     RAP. Pulse-chase labeling and immunolocalization studies of ERD2.1 and
     ERD2.2 proteins in transfected cells demonstrate a long half-life and
     Golgi localization for both receptors. Finally, overexpression of either
     ERD2.1 or ERD2.2 proteins significantly increases the capacity of cells to
     retain both ***KDEL*** and HNEL-containing proteins. Taken together,
     our results thus demonstrate that ERD2 proteins are capable of retaining
     the novel ER retention signal associated with RAP.


=> FIL STNGUIDE
COST IN U.S. DOLLARS                SINCE FILE    TOTAL
                                    ENTRY  SESSION
FULL ESTIMATED COST                   54.44    54.59

FILE 'STNGUIDE' ENTERED AT 09:47:48 ON 18 SEP 2001
USE IS SUBJECT TO THE TERMS OF YOUR CUSTOMER AGREEMENT
COPYRIGHT (C) 2001 AMERICAN CHEMICAL SOCIETY, JAPAN SCIENCE
AND TECHNOLOGY CORPORATION, AND FACHINFORMATIONSZENTRUM
KARLSRUHE

FILE CONTAINS CURRENT INFORMATION.
LAST RELOADED: Sep 14, 2001 (20010914/UP).

=> d his

     (FILE 'HOME' ENTERED AT 09:34:11 ON 18 SEP 2001)

     FILE 'BIOSIS, EMBASE, CAPLUS' ENTERED AT 09:34:29 ON 18 SEP 2001
L1          0 S LDLR354
L2          0 S LDLR 354
L3       1661 S LDLR
L4        879 S KDEL
L5        910 S KEEL
L6        263 S HDEL
L7         78 S DDEL
L8          9 S QDEL
L9         59 S ADEL
L10        16 S SDEL
L11    571052 S FUSION OR CHIMERIC OR HYBRID OR HETEROLOGOUS
L12         0 S L3 AND L4 AND L11
```

```
L13         0 S L3 AND L4
L14         0 S L3 AND L5 AND L11
L15         0 S L3 AND L6 AND L11
L16         0 S L3 AND L7
L17         0 S L3 AND L8
L18         0 S L3 AND L9
L19         0 S L3 AND L10
L20        81 S L3 AND L11
L21       195 S L4 AND L11
L22        40 S L5 AND L11
L23        60 S L6 AND L11
L24         3 S L7 AND L11
L25         3 S L8 AND L11
L26         5 S L9 AND L11
L27         1 S L10 AND L11
L28        34 DUP REM L20 (47 DUPLICATES REMOVED)
L29      9513 S LOW DENSITY LIPOPROTEIN RECEPTOR OR LDLR OR (LDLR
AND 354)
L30         3 S L29 AND L4
L31         2 DUP REM L30 (1 DUPLICATE REMOVED)

    FILE 'STNGUIDE' ENTERED AT 09:47:48 ON 18 SEP 2001

=> file biosis caplus embase
COST IN U.S. DOLLARS                 SINCE FILE     TOTAL
                                     ENTRY  SESSION
FULL ESTIMATED COST                      0.00     54.59

FILE 'BIOSIS' ENTERED AT 09:48:26 ON 18 SEP 2001
COPYRIGHT (C) 2001 BIOSIS(R)

FILE 'CAPLUS' ENTERED AT 09:48:26 ON 18 SEP 2001
USE IS SUBJECT TO THE TERMS OF YOUR STN CUSTOMER AGREEMENT.
PLEASE SEE "HELP USAGETERMS" FOR DETAILS.
COPYRIGHT (C) 2001 AMERICAN CHEMICAL SOCIETY (ACS)

FILE 'EMBASE' ENTERED AT 09:48:26 ON 18 SEP 2001
COPYRIGHT (C) 2001 Elsevier Science B.V.  All rights reserved.

=> d his

    (FILE 'HOME' ENTERED AT 09:34:11 ON 18 SEP 2001)

    FILE 'BIOSIS, EMBASE, CAPLUS' ENTERED AT 09:34:29 ON 18 SEP 2001
L1          0 S LDLR354
L2          0 S LDLR 354
L3       1661 S LDLR
L4        879 S KDEL
L5        910 S KEEL
L6        263 S HDEL
L7         78 S DDEL
L8          9 S QDEL
L9         59 S ADEL
L10        16 S SDEL
L11    571052 S FUSION OR CHIMERIC OR HYBRID OR HETEROLOGOUS
L12         0 S L3 AND L4 AND L11
L13         0 S L3 AND L4
L14         0 S L3 AND L5 AND L11
L15         0 S L3 AND L6 AND L11
L16         0 S L3 AND L7
L17         0 S L3 AND L8
L18         0 S L3 AND L9
L19         0 S L3 AND L10
L20        81 S L3 AND L11
L21       195 S L4 AND L11
L22        40 S L5 AND L11
L23        60 S L6 AND L11
L24         3 S L7 AND L11
L25         3 S L8 AND L11
L26         5 S L9 AND L11
L27         1 S L10 AND L11
L28        34 DUP REM L20 (47 DUPLICATES REMOVED)
L29      9513 S LOW DENSITY LIPOPROTEIN RECEPTOR OR LDLR OR (LDLR
AND 354)
L30         3 S L29 AND L4
L31         2 DUP REM L30 (1 DUPLICATE REMOVED)

    FILE 'STNGUIDE' ENTERED AT 09:47:48 ON 18 SEP 2001

    FILE 'BIOSIS, CAPLUS, EMBASE' ENTERED AT 09:48:26 ON 18 SEP 2001

=> s l29 and l5 and l11
L32         0 L29 AND L5 AND L11

=> s l29 and l5
L33         0 L29 AND L5

=> s l29 and l6
L34         0 L29 AND L6

=> s l29 and l7
L35         0 L29 AND L7

=> s l29 and l8
L36         0 L29 AND L8

=> s l29 and l9
L37         0 L29 AND L9

=> s l29 and l10
L38         0 L29 AND L10

=> s l29 and l11
L39       322 L29 AND L11

=> dup rem l39
PROCESSING COMPLETED FOR L39
L40       168 DUP REM L39 (154 DUPLICATES REMOVED)

=> d his

    (FILE 'HOME' ENTERED AT 09:34:11 ON 18 SEP 2001)

    FILE 'BIOSIS, EMBASE, CAPLUS' ENTERED AT 09:34:29 ON 18 SEP 2001
L1          0 S LDLR354
L2          0 S LDLR 354
L3       1661 S LDLR
L4        879 S KDEL
L5        910 S KEEL
L6        263 S HDEL
L7         78 S DDEL
L8          9 S QDEL
L9         59 S ADEL
L10        16 S SDEL
L11    571052 S FUSION OR CHIMERIC OR HYBRID OR HETEROLOGOUS
L12         0 S L3 AND L4 AND L11
L13         0 S L3 AND L4
L14         0 S L3 AND L5 AND L11
L15         0 S L3 AND L6 AND L11
L16         0 S L3 AND L7
L17         0 S L3 AND L8
L18         0 S L3 AND L9
L19         0 S L3 AND L10
L20        81 S L3 AND L11
L21       195 S L4 AND L11
L22        40 S L5 AND L11
L23        60 S L6 AND L11
L24         3 S L7 AND L11
L25         3 S L8 AND L11
L26         5 S L9 AND L11
L27         1 S L10 AND L11
L28        34 DUP REM L20 (47 DUPLICATES REMOVED)
L29      9513 S LOW DENSITY LIPOPROTEIN RECEPTOR OR LDLR OR (LDLR
AND 354)
L30         3 S L29 AND L4
L31         2 DUP REM L30 (1 DUPLICATE REMOVED)

    FILE 'STNGUIDE' ENTERED AT 09:47:48 ON 18 SEP 2001

    FILE 'BIOSIS, CAPLUS, EMBASE' ENTERED AT 09:48:26 ON 18 SEP 2001
L32         0 S L29 AND L5 AND L11
L33         0 S L29 AND L5
L34         0 S L29 AND L6
L35         0 S L29 AND L7
L36         0 S L29 AND L8
L37         0 S L29 AND L9
L38         0 S L29 AND L10
L39       322 S L29 AND L11
L40       168 DUP REM L39 (154 DUPLICATES REMOVED)

=> dup rem l21
PROCESSING COMPLETED FOR L21
L41        92 DUP REM L21 (103 DUPLICATES REMOVED)

=> dup rem l22
PROCESSING COMPLETED FOR L22
L42        34 DUP REM L22 (6 DUPLICATES REMOVED)

=> dup rem l23
PROCESSING COMPLETED FOR L23
L43        25 DUP REM L23 (35 DUPLICATES REMOVED)

=> dup rem l24
PROCESSING COMPLETED FOR L24
L44         1 DUP REM L24 (2 DUPLICATES REMOVED)

=> dup rem l25
PROCESSING COMPLETED FOR L25
L45         1 DUP REM L25 (2 DUPLICATES REMOVED)

=> dup rem l26
PROCESSING COMPLETED FOR L26
L46         3 DUP REM L26 (2 DUPLICATES REMOVED)

=> dup rem l27
PROCESSING COMPLETED FOR L27
```

L47 1 DUP REM L27 (0 DUPLICATES REMOVED)

=> d bib abs l47

L47 ANSWER 1 OF 1 EMBASE COPYRIGHT 2001 ELSEVIER SCI. B.V.
AN 92170487 EMBASE
DN 1992170487
TI Plant and mammalian sorting signals for protein retention in the endoplasmic reticulum contain a conserved epitope.
AU Denecke J.; De Rycke R.; Botterman J.
CS University of Agricultural Sciences, Uppsala Genetic Centre, Department of Molecular Genetics, Box 7003,S-75007 Uppsala, Sweden
SO EMBO Journal, (1992) 11/6 (2345-2355).
ISSN: 0261-4189 CODEN: EMJODG
CY United Kingdom
DT Journal; Article
FS 029 Clinical Biochemistry
LA English
SL English
AB We studied protein sorting signals which are responsible for the retention of reticuloplasmins in the lumen of the plant endoplasmic reticulum (ER). A non-specific passenger protein, previously shown to be secreted by default, was used as a carrier for such signals. Tagging with C-terminal tetrapeptide sequences of mammalian (KDEL) and yeast (HDEL) reticuloplasmins led to effective accumulation of the protein chimeras in the lumen of the plant ER. Some single amino acid substitutions within the tetrapeptide tag (***SDEL*** , -KDDL, -KDEI and -KDEV) can cause a complete loss of its function as a retention signal, demonstrating the high specificity of the retention machinery. However, other modifications confer efficient (-RDEL) or partial (-KEEL) retention. It is also shown that the efficiency of protein retention is not significantly impaired by an increased ligand concentration in plants. The efficiently retained chimeras (-KDEL, -HDEL and -RDEL) were shown to be recognized by a monoclonal antibody directed against the C-terminus of the mammalian reticuloplasmin protein disulfide isomerase (PDI). The recognized epitope is also present in several putative reticuloplasmins in microsomal fractions of plant and mammalian cells, suggesting that the antibodies recognize an important structural determinant of the retention signal. In addition, data are discussed which support the view that upstream sequences beyond the C-terminal tetrapeptide can influence or may be part of the structure of reticuloplasmin retention signals.

=> d his

(FILE 'HOME' ENTERED AT 09:34:11 ON 18 SEP 2001)

FILE 'BIOSIS, EMBASE, CAPLUS' ENTERED AT 09:34:29 ON 18 SEP 2001
L1 0 S LDLR354
L2 0 S LDLR 354
L3 1661 S LDLR
L4 879 S KDEL
L5 910 S KEEL
L6 263 S HDEL
L7 78 S DDEL
L8 9 S QDEL
L9 59 S ADEL
L10 16 S SDEL
L11 571052 S FUSION OR CHIMERIC OR HYBRID OR HETEROLOGOUS
L12 0 S L3 AND L4 AND L11
L13 0 S L3 AND L4
L14 0 S L3 AND L5 AND L11
L15 0 S L3 AND L6 AND L11
L16 0 S L3 AND L7
L17 0 S L3 AND L8
L18 0 S L3 AND L9
L19 0 S L3 AND L10
L20 81 S L3 AND L11
L21 195 S L4 AND L11
L22 40 S L5 AND L11
L23 60 S L6 AND L11
L24 3 S L7 AND L11
L25 3 S L8 AND L11
L26 5 S L9 AND L11
L27 1 S L10 AND L11
L28 34 DUP REM L20 (47 DUPLICATES REMOVED)
L29 9513 S LOW DENSITY LIPOPROTEIN RECEPTOR OR LDLR OR (LDLR AND 354)
L30 3 S L29 AND L4
L31 2 DUP REM L30 (1 DUPLICATE REMOVED)

FILE 'STNGUIDE' ENTERED AT 09:47:48 ON 18 SEP 2001

FILE 'BIOSIS, CAPLUS, EMBASE' ENTERED AT 09:48:26 ON 18 SEP 2001
L32 0 S L29 AND L5 AND L11
L33 0 S L29 AND L5
L34 0 S L29 AND L6
L35 0 S L29 AND L7
L36 0 S L29 AND L8
L37 0 S L29 AND L9
L38 0 S L29 AND L10
L39 322 S L29 AND L11
L40 168 DUP REM L39 (154 DUPLICATES REMOVED)
L41 92 DUP REM L21 (103 DUPLICATES REMOVED)
L42 34 DUP REM L22 (6 DUPLICATES REMOVED)
L43 25 DUP REM L23 (35 DUPLICATES REMOVED)
L44 1 DUP REM L24 (2 DUPLICATES REMOVED)
L45 1 DUP REM L25 (2 DUPLICATES REMOVED)
L46 3 DUP REM L26 (2 DUPLICATES REMOVED)
L47 1 DUP REM L27 (0 DUPLICATES REMOVED)

=> d bib abs l46

L46 ANSWER 1 OF 3 EMBASE COPYRIGHT 2001 ELSEVIER SCI. B.V.
AN 94157792 EMBASE
DN 1994157792
TI The ARG4 gene of Candida albicans.
AU Hoyer L.L.; Magee B.B.; Rikkerink E.H.A.; Scherer S.
CS Human Genome Center, Lawrence Berkeley Laboratory, Cyclotron Road,Berkeley, CA 94720, United States
SO Gene, (1994) 142/2 (213-218).
ISSN: 0378-1119 CODEN: GENED6
CY Netherlands
DT Journal; Article
FS 004 Microbiology
029 Clinical Biochemistry
LA English
SL English
AB The DNA sequence of a Candida albicans genomic fragment known to complement the arginine mutation designated arg57 in strain 1006 contains an ORF of 1404 nucleotides (nt) predicting a protein of 468 amino acids (aa). Database searches indicated that the deduced protein shares 75% identity and 85% similarity with the ARG4 protein of Saccharomyces cerevisiae. Analysis of the percent aa identity between C. albicans and S. cerevisiae sequences included in available databases suggested these values are within the range expected for biosynthetic enzymes from the two organisms which share similar function. Experiments to isolate C. albicans ARG4 by complementation in an arg4 strain of S. cerevisiae yielded a plasmid (pARG4-1) with a restriction map identical to that of the sequenced clone. From these data, we conclude that the gene previously designated ARG57 is in fact ARG4 encoding the enzyme argininosuccinate lyase (ASL). These results were unexpected, since ARG57 had been localized to chromosome 7, while a mutation causing an ASL deficiency had been linked to ***adel*** , which is on chromosome R. Transformation of C. albicans strains with pARG4-1 indicated it complemented the arginine auxotrophy in strains TMSU221 and 1435, a derivative of 1006. Examination of commonly utilized C. albicans arginine auxotrophs by spheroplast ***fusion*** analysis indicated these strains comprise two complementation groups: one consisting of 1006 and TMSU221, which are arg4, and the other of A642, hOG318, hOG357, FC18-6 and WC-5-4, which possess an undefined defect in the arginine biosynthetic pathway which we designate arg100.

=> d bib abs l46 2-
YOU HAVE REQUESTED DATA FROM 2 ANSWERS - CONTINUE? Y/(N):y

L46 ANSWER 2 OF 3 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 1
AN 1992:280227 BIOSIS
DN BA94:4877
TI ANALYSIS OF THE BiP GENE AND IDENTIFICATION OF AN ER RETENTION SIGNAL IN SCHIZOSACCHAROMYCES-POMBE.
AU PIDOUX A L; ARMSTRONG J
CS MEMBRANE MOL. BIOL. LAB., IMPERIAL CANCER RES. FUND, BOX 123, LINCOLN'S INN FIELDS, LONDON, WC2A 3PX, UK.
SO EMBO (EUR MOL BIOL ORGAN) J, (1992) 11 (4), 1583-1591.
CODEN: EMJODG. ISSN: 0261-4189.
FS BA; OLD
LA English
AB We have cloned the gene for the resident luminal ER protein BiP from the fission yeast, Schizosaccharomyces pombe. The predicted protein product is equally divergent from the budding yeast and mammalian homologues. Disruption of the BiP gene in S. pombe is lethal and BiP mRNA levels are regulated by a variety of stresses including heat shock. Immunofluorescence of cells expressing an epitope-tagged BiP protein show it to be localized to the nuclear envelope, around the cell periphery and in a reticular structure through the cytoplasm. Unexpectedly, we find the BiP protein contains an N-linked glycosylation site which can be utilized. The C-terminal four amino acids of BiP are Ala-Asp-Glu-Leu, a new variant of the XDEL sequence found at the C-termini of luminal endoplasmic reticulum proteins. To determine whether this sequence acts as a sorting signal in S. pombe we expressed an acid phosphatase ***fusion*** protein extended at its C-terminus with the amino acids ***ADEL*** . Analysis of the sorting of this ***fusion*** protein indicates that the ***ADEL*** sequence is sufficient to cause the retention of proteins in the endoplasmic reticulum. The sequences DDEL, HDEL and KDEL can also direct ER-retention of acid phosphatase in S. pombe.

L46 ANSWER 3 OF 3 BIOSIS COPYRIGHT 2001 BIOSIS
AN 1978:202058 BIOSIS
DN BA66:14555
TI GENETIC ANALYSIS OF PRODUCTS OF PROTOPLAST ***FUSION*** IN SACCHAROMYCES-CEREVISIAE.
AU GUNGE N; TAMARU A

CS CENT. RES. LAB., MITSUBISHI CHEM. IND., 1000 KAMOSHIDA, MIDORI, YOKOHAMA
227, KANAGAWA, JPN.
SO JPN J GENET, (1978) 53 (1), 41-50.
CODEN: IDZAAW. ISSN: 0021-504X.
FS BA; OLD
LA English
AB Protoplasts of S. cerevisiae were prepared from 2 different haploid strains both of mating type a, which carried different nuclear (***adel*** , ural, his4, leu2 and thr4) and mitochondrial (.rho., .omega., CR, ER and OR) markers, and were fused with the aid of polyethylene glycol. Cells of fused products (prototrophs) displayed phenotype of mating type a and were crossed to mating type .alpha./.alpha. diploids having auxotrophic markers, e.g., trp1. On sporulation of the resulting ***hybrid*** clones, as a rule, there were 3 tetrad types for mating types, i.e., 4 non-maters, 2a:2.alpha. and a:.alpha.:2 non-maters. The relative frequencies of these 3 tetrad types were close to the ones theoretically predicted from a/a/.alpha./.alpha. tetraploids, suggesting that the ***fusion*** products were a/a diploids. Auxotrophic markers involved in these crosses, which were located on 4 different chromosomes, were also segregated to yield the tetrad distributions expected from the parentages. The protoplast ***fusion*** proceeded to karyogamy to produce stable diploids. A study of mitochondrial recombination demonstrated that the ***fusion*** products accepted the mitochondrial genome (the polar gene .omega. as well as the drug resistance genes) from 1 parent of .rho.+, but not from another of neutral petite.

=> d his

(FILE 'HOME' ENTERED AT 09:34:11 ON 18 SEP 2001)

FILE 'BIOSIS, EMBASE, CAPLUS' ENTERED AT 09:34:29 ON 18 SEP 2001
L1 0 S LDLR354
L2 0 S LDLR 354
L3 1661 S LDLR
L4 879 S KDEL
L5 910 S KEEL
L6 263 S HDEL
L7 78 S DDEL
L8 9 S QDEL
L9 59 S ADEL
L10 16 S SDEL
L11 571052 S FUSION OR CHIMERIC OR HYBRID OR HETEROLOGOUS
L12 0 S L3 AND L4 AND L11
L13 0 S L3 AND L4
L14 0 S L3 AND L5 AND L11
L15 0 S L3 AND L6 AND L11
L16 0 S L3 AND L7
L17 0 S L3 AND L8
L18 0 S L3 AND L9
L19 0 S L3 AND L10
L20 81 S L3 AND L11
L21 195 S L4 AND L11
L22 40 S L5 AND L11
L23 60 S L6 AND L11
L24 3 S L7 AND L11
L25 3 S L8 AND L11
L26 5 S L9 AND L11
L27 1 S L10 AND L11
L28 34 DUP REM L20 (47 DUPLICATES REMOVED)
L29 9513 S LOW DENSITY LIPOPROTEIN RECEPTOR OR LDLR OR (LDLR AND 354)
L30 3 S L29 AND L4
L31 2 DUP REM L30 (1 DUPLICATE REMOVED)

FILE 'STNGUIDE' ENTERED AT 09:47:48 ON 18 SEP 2001

FILE 'BIOSIS, CAPLUS, EMBASE' ENTERED AT 09:48:26 ON 18 SEP 2001
L32 0 S L29 AND L5 AND L11
L33 0 S L29 AND L5
L34 0 S L29 AND L6
L35 0 S L29 AND L7
L36 0 S L29 AND L8
L37 0 S L29 AND L9
L38 0 S L29 AND L10
L39 322 S L29 AND L11
L40 168 DUP REM L39 (154 DUPLICATES REMOVED)
L41 92 DUP REM L21 (103 DUPLICATES REMOVED)
L42 34 DUP REM L22 (6 DUPLICATES REMOVED)
L43 25 DUP REM L23 (35 DUPLICATES REMOVED)
L44 1 DUP REM L24 (2 DUPLICATES REMOVED)
L45 1 DUP REM L25 (2 DUPLICATES REMOVED)
L46 3 DUP REM L26 (2 DUPLICATES REMOVED)
L47 1 DUP REM L27 (0 DUPLICATES REMOVED)

=> d bib abs l45

L45 ANSWER 1 OF 1 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 1
AN 1993:52610 BIOSIS
DN PREV199395028912
TI ***Fusion*** proteins containing the A2 domain of cholera toxin assemble with B polypeptides of cholera toxin to form immunoreactive and functional holotoxin-like chimeras.
AU Jobling, Michael G.; Holmes, Randall K. (1)
CS (1) Dep. Microbiol., Uniformed Serv. Univ. Health Sci., 4301 Jones Bridge Rd., Bethesda, Md. 20814
SO Infection and Immunity, (1992) Vol. 60, No. 11, pp. 4915-4924.
ISSN: 0019-9567.
DT Article
LA English
AB Cholera enterotoxin (CT) is produced by Vibrio cholerae and excreted into the culture medium as an extracellular protein. CT consists of one A polypeptide and five B polypeptides associated by noncovalent bonds, and CT-B interacts with CT-A primarily via the A2 domain. Treatment of CT with trypsin cleaves CT-A into A1 and A2 fragments that are linked by a disulfide bond. CT-B binds to ganglioside G-M1, which functions as the plasma membrane receptor for CT, and the enzymatic activity of A1 causes the toxic effects of CT on target cells. We constructed translational fusions that joined foreign proteins via their carboxyl termini to the A2 domain of CT-A, and we studied the interactions of the ***fusion*** poteins with CT-B. The A2 domain was necessary and sufficient to enable bacterial alkaline phosphatase (BAP), maltose-binding protein (MBP) or beta-lactamase (BLA) to associate with CT-B to form stable, immunoreactivity, holotoxin-like chimeras. Each holotoxin-like chimera was able to bind to ganglioside G-M1. Holotoxin-like chimeras containing the BAP-A2 and BLA-A2 ***fusion*** proteins had BAP activity and BLA activity, respectively. We constructed BAP-A2 mutants with altered carboxyl-terminal sequences and tested their ability to assemble into holotoxin-like chimeras. Although the carboxyl-terminal ***QDEL*** sequence of the BAP-A2 ***fusion*** protein was not required for interaction with CT-B, most BAP-A2 mutants with altered carboxyl termini did not form biotoxin-like chimeras. When holotoxin-like chimeras containing BAP-A2, MBP-A2, or BLA-A2 were synthesized in V. cholerae, they were found predominantly in the periplasm. The toxin secretory apparatus of V. cholerae was not able, therefore, to translocate these holotoxin-like chimeras across the outer membrane.

=> d bib abs l44

L44 ANSWER 1 OF 1 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 1
AN 1992:280227 BIOSIS
DN BA94:4877
TI ANALYSIS OF THE BIP GENE AND IDENTIFICATION OF AN ER RETENTION SIGNAL IN SCHIZOSACCHAROMYCES-POMBE.
AU PIDOUX A L; ARMSTRONG J
CS MEMBRANE MOL. BIOL. LAB., IMPERIAL CANCER RES. FUND, BOX 123, LINCOLN'S INN FIELDS, LONDON, WC2A 3PX, UK.
SO EMBO (EUR MOL BIOL ORGAN) J, (1992) 11 (4), 1583-1591.
CODEN: EMJODG. ISSN: 0261-4189.
FS BA; OLD
LA English
AB We have cloned the gene for the resident luminal ER protein BiP from the fission yeast, Schizosaccharomyces pombe. The predicted protein product is equally divergent from the budding yeast and mammalian homologues. Disruption of the BiP gene in S. pombe is lethal and BiP mRNA levels are regulated by a variety of stresses including heat shock. Immunofluorescence of cells expressing an epitope-tagged BiP protein show it to be localized to the nuclear envelope, around the cell periphery and in a reticular structure through the cytoplasm. Unexpectedly, we find the BiP protein contains an N-linked glycosylation site which can be utilized. The C-terminal four amino acids of BiP are Ala-Asp-Glu-Leu, a new variant of the XDEL sequence found at the C-termini of luminal endoplasmic reticulum proteins. To determine whether this sequence acts as a sorting signal in S. pombe we expressed an acid phosphatase ***fusion*** protein extended at its C-terminus with the amino acids ADEL. Analysis of the sorting of this ***fusion*** protein indicates that the ADEL sequence is sufficient to cause the retention of proteins in the endoplasmic reticulum. The sequences ***DDEL*** , HDEL and KDEL can also direct ER-retention of acid phosphatase in S. pombe.

=> d bib abs l43

L43 ANSWER 1 OF 25 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 1
AN 2000:345984 BIOSIS
DN PREV200000345984
TI Unique catalytic and molecular properties of hydrolases from Aspergillus used in Japanese bioindustries.
AU Ichishima, Eiji (1)
CS (1) Department of Bioengineering, Graduate School of Engineering, Soka University, Hachioji, Tokyo, 192-8577 Japan
SO Bioscience Biotechnology and Biochemistry, (April, 2000) Vol. 64, No. 4, pp. 675-688. print.
ISSN: 0916-8451.
DT General Review
LA English
SL English
AB This review covers the unique catalytic and molecular properties of three proteolytic enzymes and a glycosidase from Aspergillus. An aspartic proteinase from A. saitoi, aspergillopepsin I (EC 3.4.23.18), favors hydrophobic amino acids at P1 and P1' like gastric pepsin. However,

aspergillopepsin I accommodates a Lys residue at P1, which leads to activation of trypsinogens like duodenum enteropeptidase. Substitution of Asp76 to Ser or Thr and deletion of Ser78, corresponding to the mammalian aspartic proteinases, cathepsin D and pepsin, caused drastic decreases in the activities towards substrates containing a basic amino acid residue at P1. In addition, the double mutant T77D/G78(S)G79 of porcine pepsin was able to activate bovine trypsinogen to trypsin by the selective cleavage of the K6-I7 bond of trypsinogen. Deuterolysin (EC 3.4.24.39) from A. oryzae, which contains 1 g atom of zinc/mol of enzyme, is a single chain of 177 amino acid residues, includes three disulfide bonds, and has a molecular mass of 19,018 Da. It was concluded that His128, His132, and Asp164 provide the Zn2+ ligands of the enzyme according to a 65Zn binding assay. Deuterolysin is a member of a family of metalloendopeptidases with a new zinc-binding motif, aspzincin, defined by the "HEXXH + D" motif and an aspartic acid as the third zinc ligand. Acid carboxypeptidase (EC 3.4.16.1) from A. saitoi is a glycoprotein that contains both N- and O-linked sugar chains. Site-directed mutagenesis of the cpdS, cDNA encoding A. saitoi carboxypeptidase, was cloned and expressed. A. saitoi carboxypeptidase indicated that Ser153, Asp357, and His436 residues were essential for the enzymic catalysis. The N-glycanase released high-mannose type oligosaccharides that were separated on HPLC. Two, which had unique structures of Man10GlcNAc2 and Man11GlcNAc2, were characterized. An acidic
1,2-alpha-mannosidase (EC 3.2.1.113) was isolated from the culture of A. saitoi. A highly efficient overexpression system of 1,2-alpha-mannosidase ***fusion*** gene (f-msdS) in A. oryzae was made. A yeast mutant capable of producing Man5GlcNAc2 human-compatible sugar chains on glycoproteins was constructed. An expression vector for 1,2-alpha-mannosidase with the " ***HDEL*** " endoplasmic reticulum retention/retrieval tag was designed and expressed in Saccharomyces cerevisiae. The first report of production of human-compatible high mannose-type (Man5GlcNAc2) sugar chains in S. cerevisiae was described.

=> d bib abs l42 1-
YOU HAVE REQUESTED DATA FROM 34 ANSWERS - CONTINUE? Y/(N):y

L42 ANSWER 1 OF 34 EMBASE COPYRIGHT 2001 ELSEVIER SCI. B.V.DUPLICATE 1
AN 1999404788 EMBASE
TI Neural tube closure in Xenopus laevis involves medial migration, directed protrusive activity, cell intercalation and convergent extension.
AU Davidson L.A.; Keller R.E.
CS L.A. Davidson, Department of Biology, Gilmer Hall, University of Virginia, Charlottesville, VA 22903, United States
SO Development, (1999) 126/20 (4547-4556).
Refs: 46
ISSN: 0950-1991 CODEN: DEVPED
CY United Kingdom
DT Journal; Article
FS 001 Anatomy, Anthropology, Embryology and Histology
021 Developmental Biology and Teratology
LA English
SL English
AB We have characterized the cell movements and prospective cell identities as neural folds fuse during neural tube formation in Xenopus laevis. A newly developed whole-mount, two-color fluorescent RNA in situ hybridization method, visualized with confocal microscopy, shows that the dorsal neural tube gene xpax3 and the neural-crest-specific gene xslug are expressed far lateral to the medial site of neural fold ***fusion*** and that expression moves medially after ***fusion*** . To determine whether cell movements or dynamic changes in gene expression are responsible, we used low-light videomicroscopy followed by fluorescent in situ and confocal microscope. These methods revealed that populations of prospective neural crest and dorsal neural tube cells near the lateral margin of the neural plate at the start of neurulation move to the dorsal midline using distinctive forms of motility. Before fold ***fusion*** , superficial neural cells apically contract, roll the neural plate into a trough and appear to pull the superficial epidermal cell sheet medially. After neural fold ***fusion*** , lateral deep neural cells move medially by radially intercalating between other neural cells using two types of motility. The neural crest cells migrate as individual cells toward the dorsal midline using medially directed monopolar protrusions. These movements combine the two lateral populations of neural crest into a single medial population that form the roof of the neural tube. The remaining cells of the dorsal neural tube extend protrusions both medially and laterally bringing about radial intercalation of deep and superficial cells to form a single-cell-layered, pseudostratified neural tube. While ours is the first description of medially directed cell migration during neural fold ***fusion*** and re-establishment of the neural tube, these complex cell behaviors may be involved during cavitation of the zebrafish neural ***keel*** and secondary neurulation in the posterior axis of chicken and mouse. Time-lapse sequences online: http://www.people.virginia.edu/.apprx.lad4x/tubeclosure.html and http://www.biologists.com/Development/movies/dev6419.html.

L42 ANSWER 2 OF 34 EMBASE COPYRIGHT 2001 ELSEVIER SCI. B.V.
AN 1999314494 EMBASE
TI [Current status and perspectives of shoulder replacement]. AKTUELLER ENTWICKLUNGSSTAND UND PERSPEKTIVEN DER SCHULTERENDOPROTHETIK.
AU Habermeyer P.; Ebert T.
CS Dr. P. Habermeyer, ATOS-Klinik Heidelberg, Bismarckstrasse 9-15, D-69115 Heidelberg, Germany
SO Unfallchirurg, (1999) 102/9 (668-683).
ISSN: 0177-5537 CODEN: UNFAE2
CY Germany
DT Journal; General Review
FS 027 Biophysics, Bioengineering and Medical Instrumentation
033 Orthopedic Surgery
LA German
SL English; German
AB Basis of the modern shoulder implants is the Neer II-system, a non constrained total shoulder prosthesis with conforming radii of curvature and improved protection against dislocation. The second generation of shoulder prosthesis is based on the geometric shaft design of the Neer II prosthesis and offers not only a variety of modular head- and shaft-sizes but also through different radii a physiologic rotation-translation-mechanism. The third generation of humeral head prosthesis carries the concept of an anatomic reconstruction one step further and enables the surgeon to adjust the inclination and the eccentric offset of the humeral head to restore the centre of rotation. The latest development in shoulder arthroplasty are humeral head prosthesis with a fully variable 3-dimensional modularity to independently adjust the prosthestic head position regardless of the individual shaft geometry. This achieves a 3-dimensional adaptability of the prosthetic head about the stem axis in the coronary and in the sagittal plane. Besides of the humeral shaft prosthesis an alternative concept of shoulder joint replacement is established - the replacement of the humeral head articular surface. A hemispheric surface prosthesis - cup arthroplasty - is cemented onto the residual humeral head, which eliminates the obligatory humeral head resection and the reaming of the medullary canal. Bipolar shoulder prosthesis are humeral shaft prosthesis with a bi-rotational head system. Their indication is limited to pre-existing lesions of the rotator cuff and/or the glenoid surface. The inverse total shoulder prosthesis reverses the articular surface morphology of the humeral head and the glenoid. The hemispheric glenoid component serves as the centre of rotation for the concave epiphyseal proximal humerus component. This implant is especially used in cases of massive rotator cuff deficiencies. The role of shoulder prosthesis in treating acute humeral head fractures needs special consideration. A fracture prosthesis has to restore the exact length of the humerus, the centre of rotation, and the anatomical retroversion. Positioning of the tubercula and their adequate osteosynthesis is most critical and fundamental to ensure a correct healing process. A failed consolidation of the tubercula does not lead to a satisfying result. The shoulder joint replacement can be sufficiently fixated in cemented, cementless or ***hybrid*** techniques. Today several component design variations of cemented glenoid implants exist. Their main distinction is the fixation system which can be divided into two main groups - the ***keel*** - and the peg-shaped glenoid components. The peg-shaped anchorage system shall guarantee a greater stability against shear-forces. Cementless glenoid components consist of a polyethylene inlay and a surface treated metal-back with an integrated fixation system. These fixation systems are object of intensive biomechanical research and range from conventional screw fixation to specialised cone systems and self-cutting cage-screw-systems. The critical area of cementless glenoid components is the transition zone of the PE-inlay and the metal-back because of high force development. The question of implanting a hemi- or total shoulder prosthesis is answered by the morphologic changes of the glenoid articular surface, which includes the size of the subchondral defect and the underlying etiology of the shoulder joint disease, and the age of the patient. Preoperative planning must consist of an adequate radiologic work-up - X-ray, CT or MRI- to accurately assess the glenoid morphology. G. Walch categorised the different glenoid lesions and developed a very important classification of possible glenoid deformations. To corn pare and evaluate the operative results one must consider the different shoulder prosthesis and the discrepancies between a hemi- and a total shoulder prosthetic replacement. Looking at the loosening and survival rate of the implant the results are related to the type of prosthesis and the pre- operative diagnosis. The Neer total shoulder prosthesis has a 15 year survival rate of 87 %, compared to 74 % of the hemi-prosthesis. The objective for the future has to be to further advance the development of prosthetic components, especially for primary joint replacement in acute humeral head fractures. Another point of interest is how to reduce the still existing high loosening rates of the glenoid components. A fairly new research-field is the computer- assisted surgery, e.g. navigation systems and robotics. The computer-assisted navigation could be of great advantage to accurately find the individual resection plane (inclination and retroversion) of the humeral head. The use of a surgery-robot could be very helpful to reproducibly achieve the desired conformity of the articular surface when preparing the glenoid.

L42 ANSWER 3 OF 34 BIOSIS COPYRIGHT 2001 BIOSIS
AN 1999:238300 BIOSIS
DN PREV199900238300
TI ***Keel*** flowers of the Polygalaceae and Fabaceae: A functional comparison.
AU Westerkamp, C. (1); Weber, A.
CS (1) Annagraben 83, D-53111, Bonn Germany
SO Botanical Journal of the Linnean Society, (March, 1999) Vol. 129, No. 3, pp. 207-221.
ISSN: 0024-4074.
DT Article
LA English
SL English
AB Although the superficial similarity between Polygalaceae and Fabaceae

flowers is well known, a comparison between their recently more precisely defined ' ***keel*** flowers' reveals a wealth of functional congruences with regard to visual attraction, flower mechanics (interplay of fixed and mobile floral parts, presence of special contrivances such as tongue guide, foot handles, pollen cache, etc.), pollen presentation and nectar storage. Although ***keel*** flowers are principally addressed to bees, the increasingly pronounced ***fusion*** of floral parts gave rise to tubular flowers and has widened the spectrum of pollinators in the Polygalaceae. The intimate functional correspondence does not affect the assessment that the floral architecture (in terms of homologies) is quite different between the families. This is discussed with reference to the sistergroup relationship of the two families emanating from molecular systematics.

L42 ANSWER 4 OF 34 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 2
AN 1999:517278 BIOSIS
DN PREV199900517278
TI Rigid occipitocervical ***fusion*** .
AU Vale, Fernando L.; Oliver, Mark; Cahill, David W. (1)
CS (1) 4 Columbia Drive, Suite 730, Tampa, FL, 33606 USA
SO Journal of Neurosurgery, (Oct., 1999) Vol. 91, No. 2 SUPPL., pp. 144-150.
ISSN: 0022-3085.
DT Article
LA English
SL English
AB Object: Despite 50 years of neurosurgical experience, occipitocervical ***fusion*** continues to present a technical challenge to the surgeon. Traditional nonrigid techniques applied in the occiput and cervical spine often fail secondary to postsurgical cranial settling or rotational deformity. Unlike widely used nonrigid and semirigid techniques, rigid fixation of the craniocervical junction should allow correction of deformity in any plane, provide immediate stability without need for external orthosis, and prevent cranial settling. Methods: Since 1992, the senior author (D.W.C.) has used a rigid plate and screw fixation system for occipitocervical fusions. The technique proved to be more difficult than expected, and the procedure has evolved as experience was gained. The authors present a series of 24 patients and a technique that now involves the use of a custom-designed T-plate that is attached to the midline occipital " ***keel*** " at one end and to the spine at the other end by means of screw-fixed plates. Conclusions: Although it is still evolving, the current technique for obtaining rigid occipitocervical fixation allows for immediate rigidity and stability of the spine without the use of an external orthosis (that is, in the absence of osteoporosis), may be extended to any level of the spine, may be used in the absence of posterior elements, prevents postsurgical cranial settling and restenosis, facilitates reduction of the spinal deformity in any plane, and sometimes eliminates the need for an anterior (transoral) decompressive procedure.

L42 ANSWER 5 OF 34 BIOSIS COPYRIGHT 2001 BIOSIS
AN 2000:415305 BIOSIS
DN PREV200000415305
TI The anatomy of ferns: Identification and evolution.
AU Menzel, Florian (1)
CS (1) Klosterstr. 15, D-73547, Lorch Germany
SO Jahreshefte der Gesellschaft fuer Naturkunde in Wuerttemberg, (15 Dezember, 1999) Vol. 155, pp. 107-133. print.
ISSN: 0368-2307.
DT Article
LA German
SL English; German
AB No other plant taxon shows such a diversity in its vascular systems as ferns. The rachis steles of different species form one C-, X- or V-shaped bundle, two elliptical bundles, different patterns of several small bundles, etc. This diversity was used here to work out a key for the middle European pteridophytes which is based only on anatomic (i.e. histologic) characters. In most cases the species (or, at least, the genus) can be identified by one cross-section through the rachis (ferns) and the stem (lycopods and sphenophytes) respectively. This is especially useful for identifying young and sterile plants or hybrids. Due to its diversity, the stelar system can also be important for taxonomic observations. To support that an attempt was made to work out a proposal for the stelar evolution of the Polypodiales rachises. According to this theory the steles of all leptosporangiate fern species can be derived from one primitive form (fig. 33a) by processes of ***fusion*** and reduction. There is also one stele that may represent the "basic stele" of the derived ferns (fig. 27a). In the theory described, the selection pressure on the steles for mechanical rigidity is especially considered as its severity differs with different leaf sizes. Different steles among closely related species can often be explained by differences in size (e.g. in Bolbitis). This correlation can also be shown in the Aspleniaceae. All Aspleniaceae species have two small bundles in the petiole which fuse to a single one in the upper part of the leaf (fig. 23, 4, 5). Apart from the Polypodiaceae (which have side bundles, see below), the Aspleniaceae are the only family where the bundles and their xylems fuse in the middle and thus form an elliptic bundle (with an X-shaped xylem) instead of a U-shaped one. In smaller species (up to 20 cm leaf size), the single elliptic bundle occurs all over the petiole and rachis. This bundle is much less firm than the U-shaped one that occurs in other families, which could correlate to the fact that most of the members of the Aspleniaceae do not exceed approximately 30 cm in leaf size. The only very large species is Asplenium nidus (up to more than 150 cm leaf size, fig. 134). During the evolution, its stele has been modified, obviously to provide enough rigidity: The upper arms of the X-shaped xylem and bundle are strongly eked out, probably by concaulescence of the pinna strands. Besides, the rachis of this species has a significant ***keel*** which is supposed to stabilize the leaf additionally. A special case is represented by the so-called "side bundles". These are several small bundles which show a circular or semi-circular arrangement in cross-section and occur only among the derived ferns (fig. 22, 25, 26). Several characteristics make it seem improbable that they have evolved in the same way as the other bundles did. One possibility for their evolution is that they have originated from adventitious roots that fused with the rachis. These roots have only one vascular bundle and are formed multitudinously at the base of the leaf stalks among many fern species. This ***fusion*** may have provided the opportunity for leaves growing larger. This hypothesis also accounts for the side bundles only occuring among rather large species.

L42 ANSWER 6 OF 34 BIOSIS COPYRIGHT 2001 BIOSIS
AN 1999:172018 BIOSIS
DN PREV199900172018
TI Effects on plumage condition, health and mortality of dietary oats/wheat ratios to three hybrids of laying hens in different housing systems.
AU Wahlstrom, Annsofie (1); Tauson, Ragnar (1); Elwinger, Klas (1)
CS (1) Department of Animal Nutrition and Management, Swedish University of Agricultural Sciences, S-755 97, Uppsala Sweden
SO Acta Agriculturae Scandinavica Section A Animal Science, (Nov., 1998) Vol. 48, No. 4, pp. 250-259.
ISSN: 0906-4702.
DT Article
LA English
AB The effects on plumage condition and health when feeding diets with varying oats/wheat ratios to different non-beak-trimmed hybrids housed in various systems were studied in two experiments. In experiment 1 (Expt. 1) 1146 Lohmann Selected Leghorn (LSL) and 1006 Lohmann Brown (LB) birds were housed in eight aviary pens; four in each of the systems Lovsta (L) and Marielund (M), or in six groups of 24 conventional cages each (C). Two diets with a high proportion of either oats or wheat were used. Experiment 2 (Expt. 2) included 1740 LSL and 1632 SLU-1329 birds housed in 6 pens each of system M. Diets with varying proportions of oats and wheat were given. In Expt. 1, LB hens housed in C showed better plumage condition compared with those housed in the aviaries, whereas LSL birds showed the opposite trend. Housing system affected most health traits, showing superior results for system C regarding, for example, bumble foot, cleanliness of feet and ***keel*** bone lesions. In Expt. 2, feather cover deteriorated in LSL birds when the oats/wheat ratio was decreased but no such effect was found in the SLU-1329 birds.

L42 ANSWER 7 OF 34 BIOSIS COPYRIGHT 2001 BIOSIS
AN 1998:28615 BIOSIS
DN PREV199800028615
TI The occipital torus and developmental age of Sangiran-3.
AU Anton, Susan C. (1); Franzen, Jens Lorenz
CS (1) Univ. Fla., Gainesville, FL 32611 USA
SO Journal of Human Evolution, (Nov., 1997) Vol. 33, No. 5, pp. 599-610.
ISSN: 0047-2484.
DT Article
LA English
AB Since its discovery in 1938 Sangiran-3 has been considered a juvenile Pithecanthropus (Homo) erectus, and therefore, excluded from studies of adult H. erectus. Although morphological features align Sangiran-3 with H. erectus, its age designation rests on an unconvincing reconstruction of the occipital torus and lack of sutural ***fusion*** . Evaluation of the occipital shows the original reconstruction is faulty and that the current midline occipital torus is actually the right lateral torus. The new reconstruction of Sangiran-3 results in midline total morphology and development that is comparable with that in Sangiran-2. Compared with juvenile and adult H. erectus and Homo sapiens Sangiran-3 has three fully developed layers of vault bone with localized hypertrophy of the outer table into a sagittal ***keel*** , bregmatic eminence, and occipital torus. Sangiran-3's absolute vault thickness is also within the range of adult H. erectus. In addition, the coronal suture is fully interdigitated and sagittal sutural complexity is consistent with adult H. erectus. Sangiran-3's parietal sagittal contours are indistinguishable from adult H. erectus, whereas sagittal vault contours of juvenile H. erectus are usually more rounded than adults. These features indicate that Sangiran-3 is best considered a young adult H. erectus and should be included in metric and non-metric analyses of this taxon.

L42 ANSWER 8 OF 34 BIOSIS COPYRIGHT 2001 BIOSIS
AN 1997:38754 BIOSIS
DN PREV199799330742
TI Early development of chondrocranium in the tailed frog Ascaphus truei (Amphibia: Anura): Implications for anuran palatoquadrate homologies.
AU Reiss, John O.
CS Dep. Mol. Cell. Biol., LSS 444, Univ. Arizona, Tucson, AZ 85721 USA
SO Journal of Morphology, (1997) Vol. 231, No. 1, pp. 63-100.
ISSN: 0362-2525.
DT Article
LA English
AB Chondrocranial development in Ascaphus truei was studied by serial sectioning and graphical reconstruction. Nine stages (21-29; 9-18 mm TL) were examined. Mesodermal cells were distinguished from ectomesenchymal (neural crest derived) cells by retained yolk granules. Ectomesenchymal parts of the chondrocranium include the suprarostrals, pila preoptica,

anterior trabecula, and palatoquadrate. Mesodermal parts of the chondrocranium include the orbital cartilage, posterior trabecula, parachordal, basiotic lamina, and otic capsule. Development of the palatoquadrate is as follows. The pterygoid process first connects with the trabecula far rostrally; their ***fusion*** progresses caudally. The ascending process connects with a mesodermal bar that extends from the orbital cartilage to the otic capsule, and forms the ventral border of the dorsal trigeminal outlet. This bar is the "ascending process" of Ascaphus adults; it is a neurocranial, not palatoquadrate structure. The basal process chondrifies in an ectomesenchymal strand running from the quadrate ***keel*** to the postpalatine commissure. Later, the postpalatine commissure and basal process extend anteromedially to contact the floor of the anterior cupula of the otic capsule, creating separate foramina for the palatine and hyomandibular branches of the facial nerve. Based on these data, and on comparison with other frogs and salamanders, the anuran anterior quadratocranial commissure is homologized with the pterygoid process of salamanders, the anuran basal process (="pseudobasal" or "hyobasal" process) with the basal process of salamanders, and the anuran otic ledge with the basitrabecular process of salamanders. The extensive similarities in palatoquadrate structure and development between frogs and salamanders, and lacking in caecilians, are not phylogenetically informative. Available information on fossil outgroups suggests that some of these similarities are primitive for Lissamphibia, whereas for others the polarity is uncertain.

L42 ANSWER 9 OF 34 CAPLUS COPYRIGHT 2001 ACS
AN 1997:652312 CAPLUS
DN 127:278939
TI Modeling the crash response of composite structures
AU Johnson, A. F.; Kohlgruber, D.
CS Inst. Structures Design, German Aerospace Establishment (DLR), Stuttgart, 70569, Germany
SO J. Phys. IV (1997), 7(C3, International Conference on Mechanical and Physical Behaviour of Materials under Dynamic Loading, 5th, 1997), C3/981-C3/986
CODEN: JPICEI; ISSN: 1155-4339
PB Editions de Physique
DT Journal
LA English
AB Materials modeling and numerical simulation of the dynamic crash response of fiber reinforced composite structures are described. The application of explicit finite element anal. codes to composite aircraft structures and structural elements under low velocity impact conditions (up to 15 m/s) is outlined. Structures studied are designed to absorb crash energy and reduce seat de-acceleration pulses in aircraft sub-floor structures, and consist of an aircraft ***keel*** beam concept for an executive aircraft and ***hybrid*** carbon fiber/aramid fiber helicopter sub-floor box structures, and to carbon fiber/epoxy, webs with glass fiber and aramid fabric/epoxy modules, and laminated structures. Comparison between predicted structural response and failure modes with obsd. test results are given in each case.

L42 ANSWER 10 OF 34 BIOSIS COPYRIGHT 2001 BIOSIS
AN 1996:567118 BIOSIS
DN PREV199799296474
TI Foot and ***keel*** bone disorders in laying hens: Effects of artificial perch material and ***hybrid*** .
AU Tauson, Ragnar; Abrahamsson, Per
CS Dep. Anim. Nutr. Manage., Swedish Univ. Agric. Sci., Funbo-Lovsta Res. Cent., S-755 97 Uppsala Sweden
SO Acta Agriculturae Scandinavica Section A Animal Science, (1996) Vol. 46, No. 4, pp. 239-246.
ISSN: 0906-4702.
DT Article
LA English
AB The studies reported cover two experiments comprising 684 layers of the hybrids Dekalb XL, LSL and Shaver 288 (Expt. 1) and 744 ISA Brown and LSL layers (Expt. 2) kept in Get-away cages with 15 birds per cage, and in conventional cages with 4 birds per cage. At 35 and 55 weeks of age birds were scored for the appearance of bumble foot, toe pad hyperkeratosis, ***keel*** bone lesions, claw length, foot hygiene and hygiene of perch and cage floor (Get-away cages). Birds' use of perches was recorded by visual observation. Expt. 1 made use of a circular profile perch with flattened upper and lower surfaces made of European beech hardwood or of plastic, and Expt. 2 utilized the same hardwood perch and the same design but with a reduced diameter covered with a 4 mm rubber layer. All perches had equal exterior measurements, 38 times 33 mm. Bumble foot and ***keel*** bone lesions appeared only in Get-away cages and toe pad hyperkeratosis only in conventional cages. Scores for bumble foot were significantly different being inferior in LSL. In Expt. 1, the plastic perch resulted in more humble foot than the hardwood design. In Expt. 2 there was no significant effect of perch design on toe pad hyperkeratosis, ***keel*** bone lesions or bumble foot. Hygiene of feet was better in conventional cages than in Get-away cages. Although artificial materials were easier to keep clean than hardwood perches, it is concluded that plastic is not a suitable material because it increases the incidence of bumble foot, and that a soft rubber cover does not reduce humble foot or ***keel*** bone lesions compared with plain European beech hardwood perches of equal diameter. Significant interaction effects between ***hybrid*** and perch design/keeping system, especially regarding bumble foot and toe pad hyperkeratosis, indicate that genotypes are differently adapted to environmental designs in terms of the clinical health aspects studies.

L42 ANSWER 11 OF 34 BIOSIS COPYRIGHT 2001 BIOSIS
AN 1996:468790 BIOSIS
DN PREV199699191146
TI Effects on production, health and egg quality of varying proportions of wheat and barley in diets for two hybrids of laying hens kept in different housing systems.
AU Abrahamsson, Per; Tauson, Ragnar; Elwinger, Klas
CS Dep. Animal Nutrition Management, Swedish Univ. Agric. Sci., S-755 97 Uppsala Sweden
SO Acta Agriculturae Scandinavica Section A Animal Science, (1996) Vol. 46, No. 3, pp. 173-182.
ISSN: 0906-4702.
DT Article
LA English
AB A total of 2152 hens were fed one of two diets, with 25.0% wheat and 38.7% barley or 50.0% wheat and 13.7% barley. The hens were housed in battery cages with three hens per cage and in two aviary systems with tiered wire floors and litter-Lovsta with two tiers and Marielund with three tiers. Two hybrids were used: ISA Brown and Lohmann selected Leghorn. Production, interior and exterior egg quality, health, plumage, ***keel*** bone and foot condition were studied. The high-wheat diet resulted in inferior plumage condition owing to feather pecking, especially in the Leghorn ***hybrid*** , which in turn probably caused the higher feed consumption recorded. No other effects on production or egg quality traits were observed. Mortality, cannibalism, ***keel*** bone condition and foot condition were far more affected by housing system and ***hybrid*** than by diet. The highest mortality, mainly caused by cloacal cannibalism, was registered for ISA Brown in aviaries.

L42 ANSWER 12 OF 34 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 3
AN 1995:542034 BIOSIS
DN PREV199598556334
TI Midline signalling is required for Pax gene regulation and patterning of the eyes.
AU MacDonald, Rachel; Barth, K. Anukampa; Xu, Qiling; Holder, Nigel; Mikkola, Ingvild; Wilson, Stephen W. (1)
CS (1) Dev. Biol. Res. Centre, Randall Inst., Kings College London, 26-29 Drury Lane, London WC2B 5RL UK
SO Development (Cambridge), (1995) Vol. 121, No. 10, pp. 3267-3278.
ISSN: 0950-1991.
DT Article
LA English
AB Pax6 and Pax2 are members of the Pax family of transcription factors that are both expressed in the developing visual system of zebrafish embryos. Pax6 protein is present in all cells that form the neural retina and pigment epithelium, whereas Pax2 is located primarily in cells that will give rise to the optic stalk. In this study, we have addressed the role of midline signalling in the regulation of Pax2 and Pax6 distributions and in the subsequent morphogenesis of the eyes. Midline signalling is severely perturbed in cyclops mutant embryos resulting in an absence of ventral midline CNS tissue and ***fusion*** of the eyes. Mutant embryos ectopically express Pax6 in a bridge of tissue around the anterior pole of the neural ***keel*** in the position normally occupied by cells that form the optic stalks. In contrast, Pax2 protein is almost completely absent from this region in mutant embryos. Concomitant with the changes in Pax protein distribution, cells in the position of the optic stalks differentiate as retina. These results suggest that a signal emanating from the midline, which is absent in cyclops mutant embryos, may be required to promote Pax2 and inhibit Pax6 expression in cells destined to form the optic stalks. Sonic hedgehog (Shh also known as Vhh-1 and Hhg-1) is a midline signalling molecule that is absent from the neuroepithelium of cyclops mutant embryos at early developmental stages. To test the possibility that Shh might be able to regulate the spatial expression of Pax6 and Pax2 in the optic primordia, it was overexpressed in the developing CNS. The number of cells containing Pax2 was increased following shh overexpression and embryos developed hypertrophied optic stalk-like structures. Complimentary to the changes in Pax2 distribution, there were fewer Pax6-containing cells and pigment epithelium and neural retina were reduced. Our results suggest that Shh or a closely related signalling molecule emanating from midline tissue in the ventral forebrain either directly or indirectly induces the expression of Pax2 and inhibits the expression of Pax6 and thus may regulate the partitioning of the optic primordia into optic stalks and retinal tissue.

L42 ANSWER 13 OF 34 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 4
AN 1995:222565 BIOSIS
DN PREV199598236865
TI Importance of the glutamate residue of KDEL in increasing the cytotoxicity of Pseudomonas exotoxin derivatives and for increased binding to the KDEL receptor.
AU Kreitman, Robert J.; Pastan, Ira (1)
CS (1) Lab. Mol. Biol., Natl. Cancer Inst. Health, 9000 Rockville Pike, Bethesda, MD 20892 USA
SO Biochemical Journal, (1995) Vol. 307, No. 1, pp. 29-37.
ISSN: 0264-6021.
DT Article
LA English
AB It was previously shown that amino acids 609-613 (REDLK) at the C-terminus of Pseudomonas exotoxin (PE) are necessary for cytotoxicity, presumably by directing the toxin to the endoplasmic reticulum (ER) (Chaudhary, Jinno, FitzGerald and Pastan (1990) Proc. Natl. Acad. Sci. U.S.A. 87, 308-312). Using the anti-(interleukin 2 receptor (IL2R)) immunotoxin

anti-Tac(Fv)-PE38 (AT-PE38REDLK), it was found that removing the terminal lysine did not alter the activity, but replacing REDL with KDEL, the most common ER retention sequence, increased activity. To determine which amino acid in KDEL was responsible for the increase in activity, we tested eight C-terminal mutants of AT-PE38REDLK. Using IL2R-bearing MT-1 cells, we found that the glutamate residue of KDEL was required for high activity, as the cytotoxicity of AT-PE38 ending in KDEL, RDEL, ***KEEL*** or REEL was much greater than that of AT-PE38 ending in REDL, KEDL, RDDL or KDDL. Using freshly isolated lymphocytic leukaemia cells, AT-PE38 ending in KDEL. REEL or RDEL was more than 100-fold more cytotoxic than AT-PE38 ending in KEDL, REDL, RDDL or the native sequence REDLK. The RDEL sequence
also improved the cytotoxic activity of an interleukin 4-PE38 toxin ***fusion*** protein. Improved cytotoxic activity correlated with improved binding of the C-termini to the KDEL receptor on rat Golgi membranes. These data indicate that the glutamate residue of KDEL improves the cytotoxicity of PE by increasing binding to a sorting receptor which transports the toxin from the transreticular Golgi apparatus to the ER, where it is translocated to the cytosol and inhibits protein synthesis.

L42 ANSWER 14 OF 34 BIOSIS COPYRIGHT 2001 BIOSIS
AN 1994:271601 BIOSIS
DN PREV199497284601
TI Frequency of natural out-crossing in partially cleistogamous pigeonpea lines in diverse environments.
AU Saxena, K. B. (1); Jayasekera, S. J. B. A.; Ariyaratne, H. P.; Ariyanayagan, R. P.; Fonseka, H. H. D.
CS (1) Dep. Agric. Sri Lanka
SO Crop Science, (1994) Vol. 34, No. 3, pp. 660-662.
ISSN: 0011-183X.
DT Article
LA English
AB Natural out-crossing is the major cause of loss of varietal purity in pigeonpea (Cajanus cajan (L.) Millsp.). The frequency of natural out-crossing of partially cleistogamous mutant fines, characterized by a modified ***keel*** and filamentous anthers, was studied at two locations in Sri Lanka and three locations in India. Indeterminate growth habit and normal floral morphology were used as dominant markers and the frequency of natural out-crossing was estimated as percentage of the observed ***hybrid*** plants. Natural out-crossing in the mutant tines in Sri Lanka ranged from 0.14 to 1.33%, in comparison to 6.34 to 19.64% in the controls. In the Indian environments, natural outcrossing ranged from 0.16 to 2.67%. The mutant was highly stable over diverse environments, and may be of considerable economic importance in pigeonpea improvement and seed-production programs.

L42 ANSWER 15 OF 34 EMBASE COPYRIGHT 2001 ELSEVIER SCI. B.V.
AN 94335944 EMBASE
DN 1994335944
TI Surgical correction of metopic suture synostosis.
AU Eppley B.L.; Sadove A.M.
CS Division of Plastic Surgery, Indiana University Medical Center, 702 Barnhill Drive,Indianapolis, IN 46202-5200, United States
SO Clinics in Plastic Surgery, (1994) 21/4 (555-562).
ISSN: 0094-1298 CODEN: CPSUDA
CY United States
DT Journal; Article
FS 005 General Pathology and Pathological Anatomy
009 Surgery
LA English
SL English
AB Premature ***fusion*** of the metopic suture is an uncommon form of craniosynostosis, historically reported with an incidence of less than 10% among the various forms of craniosynostoses. Despite its infrequency, it is the most obvious deformity associated with premature ***fusion*** of a single suture with its prominent frontal ***keel*** , narrow forehead, and close-set eyes. This article discusses the timing, long-term results, and recent advances of surgical techniques.

L42 ANSWER 16 OF 34 CAPLUS COPYRIGHT 2001 ACS
AN 1994:307381 CAPLUS
DN 120:307381
TI Development of a novel lower limb prosthesis using low cost composite materials
AU Bartkus, Eric K.; Colvin, James M.; Arbogast, Robert E.
CS Ohio Willow Wood Co., Mount Sterling, OH, 43143, USA
SO J. Reinf. Plast. Compos. (1994), 13(4), 301-13
CODEN: JRPCDW; ISSN: 0731-6844
DT Journal
LA English
AB The increased use of graphite-reinforced advanced composites in prosthetics has increased both function and comfort for the amputee. However, lower income or less active amputees now have a more limited choice of affordable products. A new below-knee prosthetic system has been developed which utilizes low cost fiber-reinforced composite materials and innovative alignment methods to provide a comfortable and durable limb at an economical cost. A key to the design of the new limb is the use of high strength, long fatigue life fiber-reinforced sheet molding compds. (SMC) and high strength low cost pultruded components. A unique aspect of the system is that alignment adjustment is provided by interchanging a set of molded conical retaining sleeves which have an inner bore for securing the pylon. The inner bore is angled at incremental steps of one degree, ranging from zero to eight degrees total adjustment. The sleeves are slotted to allow the to be slipped over the pylon and compressed upon assembly. A simple alignment method is used to adjust the prosthesis for each amputee's gait. The alignment components are replaced with definitive components when assembling the finished prosthesis. The foot design incorporates flexible composite plates to provide a smooth transition from heel strike to toe-off during the gait cycle. The plates use compression molded SMC which is a ***hybrid*** of unidirectional fibers on the bottom (tensile) surface and randomly oriented fibers on the top (compressive) surface. The ***keel*** , which supports the plates, is also molded from randomly oriented SMC. A pultruded E glass fiber-reinforced vinyl ester solid rod is used as the pylon. Thorough component testing of the new limb has proven it t be durable and reliable. Fatigue testing on a custom built walking machine has shown no failures after two and one-half million cycles at a compressive load of 240 lbs. Clin. evaluations of the limb by seven test patients have resulted in no component failures. The patients have liked the function of the prosthesis and prosthetists have found the alignment and assembly methods easy to use.

L42 ANSWER 17 OF 34 BIOSIS COPYRIGHT 2001 BIOSIS
AN 1994:295987 BIOSIS
DN PREV199497308987
TI Foot and skeletal disorders in laying hens: Effects of perch design, ***hybrid*** , housing system and stocking density.
AU Tauson, Ragnar; Abrahamsson, Per
CS Dep. Anim. Nutr. and Manage., Swedish Univ. of Agric. Sci., Funbo-Lovsta Res. Stn., S-755 97 Uppsala Sweden
SO Acta Agriculturae Scandinavica Section A Animal Science, (1994) Vol. 44, No. 2, pp. 110-119.
ISSN: 0906-4702.
DT Article
LA English
AB In four experiments a total of 3660 SCWL laying hens kept in conventional cages at low and high stocking densities with and without a perch. Get-away (GA) cages and aviaries with litter (AL), were used for studies on the presence of humble foot (BF), distal toe pad hyperkeratosis (TPH), ***keel*** bone lesions (KBL) and of the breaking strength of tibia and humerus. Commercial hybrids were used: LSL (Expts. 1, 2 and 4); LSL and Shaver (Expt. 3). Only clearly observed in systems with perches, the incidence of BF and KBL was mostly affected by perch design, while BF was also strongly affected by strain and housing system. LSL showed significantly higher incidence of BF, especially in GA and AL. TPH, only found in conventional cages, was affected both by strain and stocking density, LSL hens and lower stocking density showing significantly lower incidence. Apart from welded wire net platforms, a European beech hardwood circular prototype perch with a flattened upper and lower surface seemed to combine in the best way until now low incidences of BF and KBL. Bone breaking strength was positively influenced by lower stocking density and the presence of a roost.

L42 ANSWER 18 OF 34 BIOSIS COPYRIGHT 2001 BIOSIS
AN 1993:279564 BIOSIS
DN PREV199396009789
TI Normal floral ontogeny and cool temperature-induced aberrant floral development in Glycine max (Fabaceae.
AU Crozier, Teresa Shuff; Thomas, Judith F. (1)
CS (1) Dep. Bot., N.C. State Univ., Raliegh, N.C. 27695-7612 USA
SO American Journal of Botany, (1993) Vol. 80, No. 4, pp. 429-448.
ISSN: 0002-9122.
DT Article
LA English
AB Floral onset in soybean (Glycine max cv. Ransom) is characterized by precocious initiation of axillary meristems in the axils of the most recently initiated leaf primordium. During floral transition, leaf morphology changes from trifoliolate leaf with stipules, to a three-lobed bract, to an unlobed bract. Soybean flowers initiated at 26/22 C day/night temperatures are normal, papilionaceous, and pentamerous. Sepal, petal, and stamen whorls are initiated unidirectionally from the abaxial to adaxial side of the floral apex. The median sepal is located abaxially and the median petal adaxially on the meristem. The organogeny of 'Ransom' flowers was found to be: sepals, petals, outer stamens plus carpel, inner stamens; or, sepals, petals, carpel, outer stamens, inner stamens. The outer stamen whorl and the carpel show possible overlap in time of initiation. Equalization of organ size occurs only within the stamen whorls. The sepals retain distinction in size, and the petals exhibit an inverse size to age relationship. The ***keel*** petals postgenitally fuse along part of their abaxial margins; their bases, however, remain free. Soybean flowers initiated at cool day/night temperatures of 18/14 C exhibited abnormalities and intermediate organs in all whorls. The gynoecium consisted of one to ten carpels (usually three or four), and carpel connation varied. ***Fusion*** of ***keel*** petals was often lacking, and stamen filaments fused erratically. Multiple carpellate flowers developed into multiple pods that were separate or variously connate. Intermediate type organs had characteristics only of organs in adjacent whorls. These aberrant flowers demonstrate that the floral meristem of soybean is not fixed or limited in its developmental capabilities and that it has the potential to produce alternate morphological patterns.

L42 ANSWER 19 OF 34 EMBASE COPYRIGHT 2001 ELSEVIER SCI. B.V.
AN 93059471 EMBASE
DN 1993059471
TI ***Hybrid*** total knee arthroplasty: Two- to five-year results using

the Miller-Galante prosthesis.
AU Kobs J.K.; Lachiewicz P.F.
CS Division of Orthopedic Surgery, 250 Burnett-Womack Building, University of North Carolina,Chapel Hill, NC 27599-7055, United States
SO Clinical Orthopaedics and Related Research, (1993) -/286 (78-87).
ISSN: 0009-921X CODEN: CORTBR
CY United States
DT Journal; Conference Article
FS 033 Orthopedic Surgery
LA English
SL English
AB Forty-one ' ***hybrid*** ' Miller-Galante total knee prostheses having porous- coated femoral and patellar components and a tibial component without a ***keel*** , cemented using low-viscosity technique, were implanted and prospectively evaluated for two to five years. The surgical technique was accurate, restoring the mechanical axis of the lower extremity to an average of 1.6.degree. varus. The average postoperative knee score was 90 points with 88% good or excellent results and 88% completely painless. Range of motion improved from a mean 88.degree. to a mean 108.degree.. Nonprogressive, incomplete radiolucent lines were present at the bone prosthesis interface in 27% of patellar, 15% of femoral, and 20% of tibial components. There were six patellar component fractures, four of which have been revised. These clinical and roentgenographic results support the ' ***hybrid*** ' technique for total knee arthroplasty. However, the use of the porous-coated metal-backed patellar component is not recommended.

L42 ANSWER 20 OF 34 EMBASE COPYRIGHT 2001 ELSEVIER SCI. B.V.
AN 92170487 EMBASE
DN 1992170487
TI Plant and mammalian sorting signals for protein retention in the endoplasmic reticulum contain a conserved epitope.
AU Denecke J.; De Rycke R.; Botterman J.
CS University of Agricultural Sciences, Uppsala Genetic Centre, Department of Molecular Genetics, Box 7003,S-75007 Uppsala, Sweden
SO EMBO Journal, (1992) 11/6 (2345-2355).
ISSN: 0261-4189 CODEN: EMJODG
CY United Kingdom
DT Journal; Article
FS 029 Clinical Biochemistry
LA English
SL English
AB We studied protein sorting signals which are responsible for the retention of reticuloplasmins in the lumen of the plant endoplasmic reticulum (ER). A non-specific passenger protein, previously shown to be secreted by default, was used as a carrier for such signals. Tagging with C-terminal tetrapeptide sequences of mammalian (KDEL) and yeast (HDEL) reticuloplasmins led to effective accumulation of the protein chimeras in the lumen of the plant ER. Some single amino acid substitutions within the tetrapeptide tag (SDEL, -KDDL, -KDEI and -KDEV) can cause a complete loss of its function as a retention signal, demonstrating the high specificity of the retention machinery. However, other modifications confer efficient (-RDEL) or partial (- ***KEEL***) retention. It is also shown that the efficiency of protein retention is not significantly impaired by an increased ligand concentration in plants. The efficiently retained chimeras (-KDEL, -HDEL and -RDEL) were shown to be recognized by a monoclonal antibody directed against the C-terminus of the mammalian reticuloplasmin protein disulfide isomerase (PDI). The recognized epitope is also present in several putative reticuloplasmins in microsomal fractions of plant and mammalian cells, suggesting that the antibodies recognize an important structural determinant of the retention signal. In addition, data are discussed which support the view that upstream sequences beyond the C-terminal tetrapeptide can influence or may be part of the structure of reticuloplasmin retention signals.

L42 ANSWER 21 OF 34 BIOSIS COPYRIGHT 2001 BIOSIS
AN 1992:186254 BIOSIS
DN BA93:97204
TI AN INTERGENERIC ***HYBRID*** OF A NATIVE MINNOW THE GOLDEN SHINER AND
AN EXOTIC MINNOW THE RUDD.
AU BURKHEAD N M; WILLIAMS J D
CS U.S. FISH WILDLIFE SERV., NATL. FISHERIES RES. CENT., 7920 NORTHWEST 71ST
ST., GAINESVILLE, FLA. 32606, USA.
SO TRANS AM FISH SOC, (1991) 120 (6), 781-795.
CODEN: TAFSAI. ISSN: 0002-8487.
FS BA; OLD
LA English
AB The ***hybrid*** golden shiner Notemigonus crysoleucas .times. rudd Scardinius erythrophthalmus is the first known nonsalmonid, intergeneric ***hybrid*** of an exotic species and a North American native species. The cross is also the first valid record of a viable ***hybrid*** involving the native golden shiner. Meristic and mensural characters of 30 artificially produced hybrids of male golden shiners and female rudds were analyzed. Forty-seven percent of the meristic traits exhibited character states intermediate between those of parents. Twenty-seven percent of the meristic characters were supernumerary, suggesting developmental instability of the ***hybrid*** genome. Mensural ***hybrid*** characters were significantly skewed to the golden shiner phenotype. The skewed mensural inheritance and other skewed patterns of morphological inheritance also suggest problems in canalization of the ***hybrid*** phenome or atypical patterns of dominance. All hybrids were identifiable by intermediate squamation of the cultrate abdomen: the ***keel*** was mostly scaled but exhibited a small fleshy ridge posteriorly. This minnow ***hybrid*** allows general inferences to be made about the phylogenetic affinity of the golden shiner to other cultrate cyprinids of Eurasia. The ***hybrid*** cross has important management and conservation implications for fishes in North America. The ***hybrid*** is an example of how an exotic species may negatively affect a native species.

L42 ANSWER 22 OF 34 BIOSIS COPYRIGHT 2001 BIOSIS
AN 1990:522161 BIOSIS
DN BA90:139437
TI FLORAL DEVELOPMENT IN OTTAWA AND FLOREX RED CLOVER TRIFOLIUM-PRATENSE
PAPILIONOIDEAE LEGUMINOSAE.
AU RETALLACK B; WILLISON J H M
CS DEP. BIOLOGY, DALHOUSIE UNIVERSITY, HALIFAX, NOVA SCOTIA, B3H 4J1 CANADA.
SO AM J BOT, (1990) 77 (9), 1222-1230.
CODEN: AJBOAA. ISSN: 0002-9122.
FS BA; OLD
LA English
AB Floral development in Florex and Ottawa cultivars of red clover (Trifolium pratense L.: Leguminosae) was examined by scanning electron microscopy. No differences between the two cultivars were found. The terminal inflorescence is initiated in the axial of the penultimate bract before the final bract is initiated. After initiation of the final bract, the remnant apical dome is transformed to become the least mature part of the inflorescence dome. Subsequent inflorescences are initiated laterally in basipetal sequence. Inflorescence development is zygomorphic. This leads to an unusual pattern of floret initiation, the oldest florets resting basally and proximal to the penultimate bract. Florets develop with zygomorphic symmetry, each whorl of flora organs developing unidirectionally from the abaxial side. Initiation of the adaxial organ of ech whorl is delayed until the abaxial organ of the succeeding whorl has been initiated. Thus there is overlapping development of the whorls of organs. The antepetalous stamens arise in close association with their respective petal primordia. As development proceeds, the corolla tube and the staminal tube exhibit basal zonal growth. In the mature flower, above the distal zone of ***fusion*** of the ***keel*** petals, marginal cells project and interlock producing a pollination mechanism that can be sprung by the pollinator.

L42 ANSWER 23 OF 34 EMBASE COPYRIGHT 2001 ELSEVIER SCI. B.V.
AN 89093195 EMBASE
DN 1989093195
TI Neural crest development in Xiphophorus fishes: scanning electron and light microscopic studies.
AU Sadaghiani B.; Vielkind J.R.
CS BC Cancer Research Centre, Vancouver, BC, V5Z 1L3, Canada
SO Development, (1989) 105/3 (487-504).
ISSN: 0950-1991 CODEN: DEVPED
CY United Kingdom
DT Journal
FS 001 Anatomy, Anthropology, Embryology and Histology
021 Developmental Biology and Teratology
LA English
SL English
AB We have studied neural crest development in two teleost fish species, Xiphophorus maculatus (platyfish) and X. helleri (swordtail), and found similarities to that in other vertebrates but also some important differences. Unlike in other vertebrates, segregation of neural crest cells occurs in masses or groups from the dorsal-lateral part of the neural ***keel*** (tube) except in the mesencephalon region, where neural crest cells segregate from the dorsal-midline and in the most anterior trunk region, where they segregate individually. However, the cells were found in the usual neural tube-somite and somite-ectoderm migration pathways. Notably numerous cells, presumed in part to be neural crest cells, were found in a third location, dorsally on the neural tube. These cells exhibit a series of morphological stages referred to as 'covering', 'condensation', and 'differentiation'. A great amount of ECM was observed in these fish and can be temporally and regionally correlated with the appearance of the neural crest cells. No major differences could be detected between the two fish species with the exception that segregation and appearance of neural crest cells in various locations occur earlier in the platyfish. This time difference could lead to perturbations in neural crest cell development in certain platyfish-swordtail hybrids and may contribute to the formation of neural-crest-derived pigment cell tumours, melanomas, in these hybrids.

L42 ANSWER 24 OF 34 BIOSIS COPYRIGHT 2001 BIOSIS
AN 1991:246394 BIOSIS
DN BA91:126949
TI CHROMOSOMAL VARIATION IN LOTUS-ALPINUS FABACEAE.
AU O'DONOUGHUE L S; GRANT W F
CS DEP. PLANT SCI., P.O. BOX 4000, MACDONALD COLLEGE OF MCGILL UNIV., STE.
ANNE DE BELLEVUE, QUEBEC, CANADA H9X 1C0.
SO PLANT SPECIES BIOL, (1989) 4 (2), 117-122.
CODEN: PSBIEK.
FS BA; OLD
LA English
AB An accession of Lotus alpinus Schleich. (2n=2x=12) from Turkey in which B

chromosomes have been found was studied morphologically and karyologically. Chromosome numbers were observed in 519 cells from nine plants which all exhibited mixoploidy (2n = 11, 12, 12 + 1B, 12 + 2B and over 20). ***Keel*** tip color, stem pubescence, and inflorescence size differed from a collection of this species from Switzerland. While the percentage of total lengths of the chromosome complements and the relative chromosome lengths in the two accessions were very similar, the total complement lengths differed considerably (23.14 .mu.m Turkey vs. 29.46 .mu.m Switzerland). This karyological difference is not considered tobe the result of the presence of B chromosomes, but probably the result of hybridization between different genotypes. Aborted seed pods were observed which lent credibility to this hypothesis. Plants of this acccession may have arisen as a result of hybridization between Lotus corniculatus and/or L. alpinus as both diploid and tetraploid cytotypes are reported in the Turkey collection for these species. The data would lend support for their ***hybrid*** origin.

L42 ANSWER 25 OF 34 BIOSIS COPYRIGHT 2001 BIOSIS
AN 1988:218809 BIOSIS
DN BA85:108044
TI THE CYTOGENETIC RELATIONSHIP BETWEEN CICER-JUDAICUM BOISS. AND
CICER-CHORASSANICUM BGE. M. POP.
AU AHMAD F; SLINKARD A E; SCOLES G J
CS DEP. CROP SCI. PLANT ECOL., UNIV. SASKATCHEWAN, SASKATOON, SASK., CANADA
S7N 0W0.
SO GENOME, (1987) 29 (6), 883-886.
CODEN: GENOE3. ISSN: 0831-2796.
FS BA; OLD
LA English
AB A single plant was produced of the interspecific ***hybrid*** Cicer judaicum Boiss. (2n = 17) .times. Cicer chorassanicum (Bge.) M. Pop. (2n = 16), but none was produced from the reciprocal cross. The ***hybrid*** plant was intermediate in morphology between the parental species with the dominant purple flower color of C. judaicum. The ***hybrid*** plant had a diploid somatic chromosome number of 2n = 16 and was characterized cytologically. The ***hybrid*** had a low chiasmata frequency (5.4 .+-. 1.2 vs. 12.1 and 11.4 in the parental species) per cell and was highly sterile. The flowers were abnormal in that the stigma and style grew out of the ***keel*** , while the anthers remained inside. Sterility and abnormal floral structure may play important roles in maintenance of species identity.

L42 ANSWER 26 OF 34 BIOSIS COPYRIGHT 2001 BIOSIS
AN 1988:129329 BIOSIS
DN BA85:64156
TI SOMATIC HYBRIDIZATION BETWEEN BIRDSFOOT TREFOIL LOTUS-CORNICULATUS L. AND
LOTUS-CONIMBRICENSIS WILLD.
AU WRIGHT R L; SOMERS D A; MCGRAW R L
CS DEP. AGRONOMY PLANT GENET., UNIV. MINN., 411 BORLAUG HALL, 1991 UPPER
BUFORD CIRCLE, ST. PAUL, MINN.
SO THEOR APPL GENET, (1987) 75 (1), 151-156.
CODEN: THAGA6. ISSN: 0040-5752.
FS BA; OLD
LA English
AB Somatic ***hybrid*** plants were produced by ***fusion*** of birdsfoot trefoil (Lotus corniculatus) cv 'Leo' and L. conimbricensis Willd. protoplasts. Birdsfoot trefoil etiolated hypocotyl protoplasts were inactivated with iodoacetate to inhibit cell division prior to ***fusion*** with L. conimbricensis suspension culture protoplasts. L. conimbricensis protoplasts divided to form callus which did not regenerate plants. Thus, plant regeneration from protoplast-derived callus was used to tentatively identify somatic ***hybrid*** cell lines. Plants regenerated from three cell lines exhibited additive combinations of parental isozymes of phosphoglucomutase, and L. conimbricensis-specific esterases indicating that they were somatic hybrids. The somatic chromosome number of one somatic ***hybrid*** was 36. The other somatic ***hybrid*** exhibited variable chromosome numbers ranging from 33 to 40. These observations approximate the expected combination of the birdsfoot trefoil (2n = 4.times. = 24) and L. conimbricensis (2n = 2.times. = 12) genomes. Somatic ***hybrid*** flowers were less yellow than birdsfoot trefoil flowers and had purple ***keel*** tips, a trait inherited from the white flowered L. conimbricensis. Somatic hybrids also had inflorescence structure that was intermediate to the parents. Fifteen somatic ***hybrid*** plants regenerated from the three callus lines were male sterile. Successful fertilization in backcrosses with birdsfoot trefoil pollen has not yet been obtained suggesting that the hybrids are also female sterile. This is the first example of somatic hybridization between these two sexually incompatible Lotus species.

L42 ANSWER 27 OF 34 BIOSIS COPYRIGHT 2001 BIOSIS
AN 1987:62529 BIOSIS
DN BA83:30855
TI A NEW SPECIES OF RAPHIONACME PERIPLOCACEAE FROM SOUTH WEST AFRICA-NAMIBIA.
AU VENTER H J T; VERHOEVEN R L
CS DEPARTMENT OF BOTANY, UNIVERSITY OF THE ORANGE FREE STATE, P.O. BOX 339,
BLOEMFONTEIN, 9300 REPUBLIC OF SOUTH AFRICA.
SO S AFR J BOT, (1986) 52 (4), 332-334.
CODEN: SAJBDD. ISSN: 0254-6299.
FS BA; OLD
LA English
AB Raphionacme namibiana Venter & Verhoeven, a new species from South West
Africa/Namibia is described. The species is recognised by the unusual ***fusion*** of the corona lobes to the corolla lobes, the stout ***keel*** -shaped follicles and the peculiar seed which has a marginal ring of hairs instead of the normal micropylar coma. Raphionacme namibiana is related to Raphionacme grandiflora, N.E. Br. from Tropical East and Central Africa. In both species the corona, stamens and corolla are quite alike. The two species, however, differ distinctly with regard to their fruit and seed.

L42 ANSWER 28 OF 34 BIOSIS COPYRIGHT 2001 BIOSIS
AN 1985:296710 BIOSIS
DN BA79:76706
TI AN ATTEMPT AT PREDICTING GENETIC EFFECTS IN CASE OF POSSIBLE CONTACT
BETWEEN 2 SPP. THE STICKLEBACKS PUNGITIUS-PUNGITIUS AND PUNGITIUS-PLATYGASTER AS A RESULT OF THE DISTURBANCE OF THEIR NATURAL
RANGES.
AU ZYUGANOV V V
CS N.K. KOLTSOV INST. DEV. BIOL., ACAD. SCI. USSR, MOSCOW, USSR.
SO GENETIKA, (1984) 20 (10), 1691-1700.
CODEN: GNKAA5. ISSN: 0016-6758.
FS BA; OLD
LA Russian
AB Crosses were made between 2 closely related allopatric species of Pungitius genera (Gasterosteidae, Pisces), namely, the northern species P. pungitius L. and the southern P. platygaster Kessler. The crosses were made in laboratory conditions (in aquaria) and under controlled conditions in nature (in ponds). As the ranges of the 2 spp. were disturbed and the species are expected to come in contact in the nearest future (rivers Irtysh, Volga [USSR]), potential mechanisms of reproductive isolation were studied. No well developed mechanisms of ethological isolation were found, the F1 and F2 hybrids being fertile and the ***hybrid*** population self-reproductive. By a complex of morphological characters, the hybrids are easily distinguishable from both parental species. The comparison of inheritance of the polymorphic character numbers of lateral bony plates on the body in Pungitius with that of the homologous character in the related genera Gasterosteus revealed no similarity. The character ***keel*** on the caudal peduncle is inherited similarly in the 2 genera. The results obtained predict possible introgressive hybridization in case of contact between P. pungitius and P. platygaster in natural conditions.

L42 ANSWER 29 OF 34 BIOSIS COPYRIGHT 2001 BIOSIS
AN 1985:239872 BIOSIS
DN BA79:19868
TI DORSAL AND ANAL FIN RAYS OF THE JAPANESE ANCHOVY ENGRAULIS-JAPONICA AND
THEIR PTERYGIOPHORES.
AU KINOSHITA T
CS LAB. BIOL. FISH POPULATION, FAC. FISHERIES, HOKKAIDO UNIV.
SO BULL FAC FISH HOKKAIDO UNIV, (1984) 35 (2), 66-82.
CODEN: HOSGAD. ISSN: 0018-3458.
FS BA; OLD
LA English
AB The 1st dorsal pterygiophore of the Japanese anchovy has a large median ***keel*** projecting forward. This pterygiophore is not formed by ***fusion*** of the 2 anterior proximal radials, but by its own developmental transfiguration. Although each dorsal and anal pterygiophore from the 2nd to the last was associated serially with 1 branched ray, the 1st dorsal pterygiophore supported 3 or 4 unbranched rays and the 1st anal pterygiophore 2, 3 or 4 unbranched rays. These were called 2-, 3- and 4-type in accordance with the number of rays. Among the rays supported by the 1st pterygiophore under both the dorsal and anal fins, the anterior most ray in the 3-type and the 2 anterior rays in the 4-type were identified as vestigial rays. The principal rays in the dorsal and anal fins of the Japanese anchovy consist of 2 unbranched rays succeeded by branched rays. The numbers of dorsal and anal fin rays are 15 and 18, respectively, in modes of the frequency distributions.

L42 ANSWER 30 OF 34 BIOSIS COPYRIGHT 2001 BIOSIS
AN 1982:198994 BIOSIS
DN BA73:58978
TI THE TAXONOMIC VALUE OF FLORAL CHARACTERS IN TRIBE TRIGONELLEAE LEGUMINOSAE
WITH SPECIAL REFERENCE TO MEDICAGO.
AU SMALL E; CROMPTON C W; BROOKES B S
CS BIOSYSTEMATICS RES. INST., AGRIC. CAN., OTTAWA, ONT., CAN. K1A OC6.
SO CAN J BOT, (1981) 59 (9), 1578-1598.
CODEN: CJBOAW. ISSN: 0008-4026.
FS BA; OLD
LA English
AB The legume tribe Trigonelleae comprises Medicago (with M. arborea sometimes segregated as the monotypic genus Rhodusia), Melilotus, Trigonella and the monotypic Factorovskya. The wisdom of segregating the 2 monotypic genera is questioned and many species are claimed to represent intergrading variation between Medicago and either Melilotus or Trigonella, or between the latter pair. The present numerical taxonomic

analysis (agglomerative clustering and ordination) of floral characters indicated that Medicago, Melilotus and Trigonella are distinguished on the basis of combinations of floral attributes, although no single characteristic was capable of separating them completely. Trigonella sect. Bucerates were distinctive from the remaining species of Trigonella examined. Limited evidence was found for segregating Medicago arborea as a monotypic genus. Factorovskya aschersoniana proved distinctive, but its relationships remain enigmatic. Discriminant analysis was employed to test the affinities of problematical species allegedly intermediate between Medicago, Trigonella and Melilotus. Most of the intermediate species were much closer to 1 of the genera than to the others. A syndrome of morphological features was discovered to separate the Trigonelleae into 2 classes of plants, the 1 group including Medicago, Factorovskya and Trigonella sect. Bucerates, and the other comprising Melilotus and the remaining examined species of Trigonella. The former group contrasts with the latter by possessing interlocking wing and ***keel*** petals, relatively less apical ***fusion*** of the ***keel*** petals and relatively well-developed wing petal horn; and by having a greater frequency of species with dilated filaments, with staminal tubes which are conical at the apex rather than blunt, and with standard petals having more than 3 clusters of veins. The latter 3 differences are less frequent between the 2 groups than the 1st 3. The floral syndrome reflects adaptation of the former group of plants to outcrossing (perhaps relictual adaptation in the inbreeding species) by means of the tripping mechanism which is well-known in Medicago. The taxonomic significance of the syndrome is difficult to ascertain, as it may have developed independently in the different genera in which it occurs.

L42 ANSWER 31 OF 34 BIOSIS COPYRIGHT 2001 BIOSIS
AN 1982:225281 BIOSIS
DN BA73:85265
TI PALAEOSTACHYA-DIRCEI NEW-SPECIES AN AUTHIGENICALLY CEMENTED EQUISETALEAN
STROBILUS FROM THE MIDDLE PENNSYLVANIAN OF SOUTHERN ILLINOIS USA.
AU GASTALDO R A
CS DEP. GEOL., AUBURN UNIV., AUBURN, ALA. 36849, USA.
SO AM J BOT, (1981) 68 (10), 1306-1318.
CODEN: AJBOAA. ISSN: 0002-9122.
FS BA; OLD
LA English
AB P. dircei sp. nov. is described from an authigenically cemented specimen collected from the Anna Shale Member occurring above the Herrin (No. 6) Coal Member and below the Brereton Limestone in the Carbondale Formation, Kewanee Group (Middle Pennsylvanian). The strobilus is 3-dimensionally disposed within the matrix, allowing the preparation of ground thin sections, as well as selected maceration of the specimen. The imbricate strobilus is preserved for at least 7 cm of its original length and is composed of alternating whorls of sterile bracts and fertile sporangiophores. An articulated axis extends the length of the strobilus and attains a maximum width of 3 mm at the nodal areas. A whorl of 24 sterile bracts arises at each node, with each bract emerging at a 90 angle from the axis. Bracts are free except for a slight adaxial ***fusion*** at their point of origin. A slight downward-projecting ***keel*** develops at the point where the bract begins ascending at least past the 2nd supra-adjacent node, where it is appressed into an abaxial furrow of the superposed bract. A whorl of sporangiophores originates above the bracts and is equal in number to the bracts. The sporangiophores are obliquely inserted on the axis and possess 4 superposed and thin-walled sporangia inserted upon a (?) cruciate head. Spores assigned to Calamospora were recovered and range in diameter from 68-115 .mu.m (.hivin.x = 89 .mu.m). The cone appears to be homosporous. P. dircei is compared to the reported permineralized and coalified compression species and appears similar to P. vera Seward and P. gracilis Renault.

L42 ANSWER 32 OF 34 EMBASE COPYRIGHT 2001 ELSEVIER SCI. B.V.
AN 80201758 EMBASE
DN 1980201758
TI Radiotelemetry of avian electrocardiogram.
AU Filshie J.H.; Duncan I.J.H.; Clark J.S.B.
CS Agric. Res. Counc. Poultry Res., Cent. King's Build., Edinburgh EH9 3JS, United Kingdom
SO Medical and Biological Engineering and Computing, (1980) 18/5 (633-637).
CODEN: MBECDY
CY United Kingdom
DT Journal
FS 027 Biophysics, Bioengineering and Medical Instrumentation
018 Cardiovascular Diseases and Cardiovascular Surgery
002 Physiology
LA English
AB A radiotelemetry systen has been developed which is capable of transmitting electrocardiogram signals from domestic fowl. The transmitter circuit consists of a frequency-modulated Colpitts oscillator operating at 104.5 MHz, followed by a single-ended amplifier. The devices are fabricated as thin-film ***hybrid*** microcircuits. Transmitters were encapsulated in silicone rubber and implanted subcutaneously over the pectoral muscles in domestic QRS complex, which could be used as a trigger in measuring heart rate, and a good ratio of usable to unusable trace. These results were obtained when the electrodes were sutured 60 mm apart to the connective tissue covering the ***keel*** bone. Implantation of the devices did not affect the behaviour of the birds and there were no pathological lesions associated with them up to four weeks later.

L42 ANSWER 33 OF 34 BIOSIS COPYRIGHT 2001 BIOSIS
AN 1980:245191 BIOSIS
DN BA70:37687
TI PLEISTOCENE SEMI SPECIATION IN PLATYSMA-MINUS COLEOPTERA CARABIDAE.
AU BRANDMAYR P; DRIOLI G
CS IST. ZOOL. ANAT. COMP., UNIV. TIESTE, VIA A. VALERIO 32, 34100 TRIESTE,
ITALY.
SO MEM SOC ENTOMOL ITAL, (1978 (1979)) 57 (0), 101-116.
CODEN: MSEIAW. ISSN: 0037-8747.
FS BA; OLD
LA Italian
AB Based on specimens from some European museums and collections, morphological features and their geographic variation are studied in Eurosibiric P. minus Gyll., 1827 and the closely related species P. oenotrium Ravizza, 1975, described from Italy. Internal sacs, inflated, revealed a new diagnostic character. Description of a new subspecies P. m. turcicum, from Anatolia and European Turkey is given. P. minus and P. oenotrium are contiguous along a line ranging from Brittany [France] across Jura and Carinthia as far as Yugoslavia. In the northwestern and more recent part of the contact zone hybridization takes place and clinal structure was observed in populations of the Swiss plateau. In this belt of secondary intergradation only intermediate individuals are present. In the eastern and less recent part of the boundary (especially in Carinthia, Styria and Slovenia, the rest of Yugoslavia being underworked) ***hybrid*** phenotypes are missing. No explanation can be given of this fact, but the longitudinal ***keel*** of male urosternum VII, a structure involved in mating behavior, is reduced in P. oenotrium. Divergence in this real couplet of semispecies probably arose during Pleistocene, as allopatric populations outlived glacial times in east-west disjunct refugia and an interesting comparison can be made with North American Cicindela spp. studied by Freitag (1965). In the European species of Melanius zoogeographic situation as a whole and specific amplitudes of environmental requirements agree with differential Quarternary evolution, which seems to have occurred very little in the extremely eurytopic P. nigrita only. Results are discussed also with regard to current hypotheses on quaternary evolution in Carabidae, as they are reviewed in Thiele (1977). Pleistocenic sub (semi) speciation apparently occurred also in macropterous or dimorphic hygrosylvicolous and/or eurytopic species of wet habitats with high dispersal power. A key of European species and subspecies of Melanius Bon (= Pseudomaseus Chaud) is given.

L42 ANSWER 34 OF 34 EMBASE COPYRIGHT 2001 ELSEVIER SCI. B.V.
AN 74024720 EMBASE
DN 1974024720
TI ***Fusion*** of the vocal cords following intubation and tracheostomy.
AU Kirchner J.A.; Sasaki C.T.
CS Sect. Otolaryngol., Dept. Surg., Yale Univ. Sch. Med., New Haven, Conn., United States
SO Transactions of the American Academy of Ophthalmology and Otolaryngology,
(1973) 77/2 (ORL88-ORL91).
CODEN: TAAOAF
DT Journal
FS 011 Otorhinolaryngology
007 Pediatrics and Pediatric Surgery
LA English
AB Glottic ***fusion*** is described as a complication of endotracheal intubation followed by tracheostomy. The conditions predisposing to this situation include abrasion of the glottic epithelium and prolonged tracheostomy. The appearance by mirror examination mimics that of bilateral recurrent laryngeal nerve paralysis. However, direct laryngotracheoscopy provides unequivocal evidence of vocal cord ***fusion*** as the primary cause of the laryngeal obstruction. The cessation of phasic laryngeal abductor activity as a result of the decreased ventilatory resistance from tracheostomy is implicated in the pathogenesis of this condition. Treatment may consist simply of repeated dilatations or the insertion of a McNaught ***keel*** while normal ventilatory resistance is being reestablished.

=> d his

(FILE 'HOME' ENTERED AT 09:34:11 ON 18 SEP 2001)

FILE 'BIOSIS, EMBASE, CAPLUS' ENTERED AT 09:34:29 ON 18 SEP 2001
L1 0 S LDLR354
L2 0 S LDLR 354
L3 1661 S LDLR
L4 879 S KDEL
L5 910 S KEEL
L6 263 S HDEL
L7 78 S DDEL
L8 9 S QDEL
L9 59 S ADEL
L10 16 S SDEL
L11 571052 S FUSION OR CHIMERIC OR HYBRID OR HETEROLOGOUS
L12 0 S L3 AND L4 AND L11
L13 0 S L3 AND L4
L14 0 S L3 AND L5 AND L11
L15 0 S L3 AND L6 AND L11
L16 0 S L3 AND L7

L17 0 S L3 AND L8
L18 0 S L3 AND L9
L19 0 S L3 AND L10
L20 81 S L3 AND L11
L21 195 S L4 AND L11
L22 40 S L5 AND L11
L23 60 S L6 AND L11
L24 3 S L7 AND L11
L25 3 S L8 AND L11
L26 5 S L9 AND L11
L27 1 S L10 AND L11
L28 34 DUP REM L20 (47 DUPLICATES REMOVED)
L29 9513 S LOW DENSITY LIPOPROTEIN RECEPTOR OR LDLR OR (LDLR AND 354)
L30 3 S L29 AND L4
L31 2 DUP REM L30 (1 DUPLICATE REMOVED)

FILE 'STNGUIDE' ENTERED AT 09:47:48 ON 18 SEP 2001

FILE 'BIOSIS, CAPLUS, EMBASE' ENTERED AT 09:48:26 ON 18 SEP 2001
L32 0 S L29 AND L5 AND L11
L33 0 S L29 AND L5
L34 0 S L29 AND L6
L35 0 S L29 AND L7
L36 0 S L29 AND L8
L37 0 S L29 AND L9
L38 0 S L29 AND L10
L39 322 S L29 AND L11
L40 168 DUP REM L39 (154 DUPLICATES REMOVED)
L41 92 DUP REM L21 (103 DUPLICATES REMOVED)
L42 34 DUP REM L22 (6 DUPLICATES REMOVED)
L43 25 DUP REM L23 (35 DUPLICATES REMOVED)
L44 1 DUP REM L24 (2 DUPLICATES REMOVED)
L45 1 DUP REM L25 (2 DUPLICATES REMOVED)
L46 3 DUP REM L26 (2 DUPLICATES REMOVED)
L47 1 DUP REM L27 (0 DUPLICATES REMOVED)

=> d bib abs l43 1-
YOU HAVE REQUESTED DATA FROM 25 ANSWERS - CONTINUE? Y/(N):y

L43 ANSWER 1 OF 25 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 1
AN 2000:345984 BIOSIS
DN PREV200000345984
TI Unique catalytic and molecular properties of hydrolases from Aspergillus used in Japanese bioindustries.
AU Ichishima, Eiji (1)
CS (1) Department of Bioengineering, Graduate School of Engineering, Soka University, Hachioji, Tokyo, 192-8577 Japan
SO Bioscience Biotechnology and Biochemistry, (April, 2000) Vol. 64, No. 4, pp. 675-688. print.
ISSN: 0916-8451.
DT General Review
LA English
SL English
AB This review covers the unique catalytic and molecular properties of three proteolytic enzymes and a glycosidase from Aspergillus. An aspartic proteinase from A. saitoi, aspergillopepsin I (EC 3.4.23.18), favors hydrophobic amino acids at P1 and P1' like gastric pepsin. However, aspergillopepsin I accommodates a Lys residue at P1, which leads to activation of trypsinogens like duodenum enteropeptidase. Substitution of Asp76 to Ser or Thr and deletion of Ser78, corresponding to the mammalian aspartic proteinases, cathepsin D and pepsin, caused drastic decreases in the activities towards substrates containing a basic amino acid residue at P1. In addition, the double mutant T77D/G78(S)G79 of porcine pepsin was able to activate bovine trypsinogen to trypsin by the selective cleavage of the K6-I7 bond of trypsinogen. Deuterolysin (EC 3.4.24.39) from A. oryzae, which contains 1 g atom of zinc/mol of enzyme, is a single chain of 177 amino acid residues, includes three disulfide bonds, and has a molecular mass of 19,018 Da. It was concluded that His128, His132, and Asp164 provide the Zn2+ ligands of the enzyme according to a 65Zn binding assay. Deuterolysin is a member of a family of metalloendopeptidases with a new zinc-binding motif, aspzincin, defined by the "HEXXH + D" motif and an aspartic acid as the third zinc ligand. Acid carboxypeptidase (EC 3.4.16.1) from A. saitoi is a glycoprotein that contains both N- and O-linked sugar chains. Site-directed mutagenesis of the cpdS, cDNA encoding A. saitoi carboxypeptidase, was cloned and expressed. A. saitoi carboxypeptidase indicated that Ser153, Asp357, and His436 residues were essential for the enzymic catalysis. The N-glycanase released high-mannose type oligosaccharides that were separated on HPLC. Two, which had unique structures of Man10GlcNAc2 and Man11GlcNAc2, were characterized. An acidic
1,2-alpha-mannosidase (EC 3.2.1.113) was isolated from the culture of A. saitoi. A highly efficient overexpression system of 1,2-alpha-mannosidase ***fusion*** gene (f-msdS) in A. oryzae was made. A yeast mutant capable of producing Man5GlcNAc2 human-compatible sugar chains on glycoproteins was constructed. An expression vector for 1,2-alpha-mannosidase with the " ***HDEL*** " endoplasmic reticulum retention/retrieval tag was designed and expressed in Saccharomyces cerevisiae. The first report of production of human-compatible high mannose-type (Man5GlcNAc2) sugar chains in S. cerevisiae was described.

L43 ANSWER 2 OF 25 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 2
AN 2001:200429 BIOSIS
DN PREV200100200429
TI Root hair formation: F-actin-dependent tip growth is initiated by local assembly of profilin-supported F-actin meshworks accumulated within expansin-enriched bulges.
AU Baluska, Frantisek (1); Salaj, Jan; Mathur, Jaideep; Braun, Markus; Jasper, Fred; Samaj, Josef; Chua, Nam-Hai; Barlow, Peter W.; Volkmann, Dieter
CS (1) Zellbiologie der Pflanzen, Botanisches Institut, Rheinische Friedrich-Wilhelms-Universitaet Bonn, Kirschallee 1, D-53115, Bonn: baluska@uni-bonn.de Germany
SO Developmental Biology, (November, 2000) Vol. 227, No. 2, pp. 618-632. print.
ISSN: 0012-1606.
DT Article
LA English
SL English
AB Plant root hair formation is initiated when specialized elongating root epidermis cells (trichoblasts) assemble distinct domains at the plasma membrane/cell wall cell periphery complexes facing the root surface. These localities show accumulation of expansin and progressively transform into tip-growing root hair apices. Experimentation showed that trichoblasts made devoid of microtubules (MTs) were unaffected in root hair formation, whereas those depleted of F-actin by the G-actin sequestering agent latrunculin B had their root hair formation blocked after the bulge formation stage. In accordance with this, MTs are naturally depleted from early outgrowing bulges in which dense F-actin meshworks accumulate. These F-actin caps remain associated with tips of emerging and growing root hairs. Constitutive expression of the GFP-mouse talin ***fusion*** protein in transgenic Arabidopsis, which visualizes all classes of F-actin in a noninvasive mode, allowed in vivo confirmation of the presence of distinct F-actin meshworks within outgrowing bulges and at tips of young root hairs. Profilin accumulates, at both the protein and the mRNA levels, within F-actin-enriched bulges and at tips of emerging hairs. ER-based calreticulin and ***HDEL*** proteins also accumulate within outgrowing bulges and remain enriched at tips of emerging hairs. All this suggests that installation of the actin-based tip growth machinery takes place only after expansin-associated bulge formation and requires assembly of profilin-supported dynamic F-actin meshworks.

L43 ANSWER 3 OF 25 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 3
AN 2000:240885 BIOSIS
DN PREV200000240885
TI Two distinct domains of the beta-subunit of glucosidase II interact with the catalytic alpha-subunit.
AU Arendt, Christopher W.; Ostergaard, Hanne L. (1)
CS (1) Department of Medical Microbiology and Immunology, University of Alberta, Edmonton, Alberta, T6G 2S2 Canada
SO Glycobiology, (May, 2000) Vol. 10, No. 5, pp. 487-492.
ISSN: 0959-6658.
DT Article
LA English
SL English
AB Recent purification and cDNA cloning of the endoplasmic reticulum processing enzyme glucosidase II have revealed that it is composed of two soluble proteins: a catalytic alpha-subunit and a beta-subunit of unknown function, both of which are highly conserved in mammals. Since the beta-subunit, which contains a C-terminal His-Asp-Glu-Leu (***HDEL***) motif, may function to link the catalytic subunit to the KDEL receptor as a retrieval mechanism, we sought to map the regions of the mouse beta-subunit protein responsible for mediating the association with the alpha-subunit. By screening a panel of recombinant beta-subunit glutathione S-transferase ***fusion*** proteins for the ability to precipitate glucosidase II activity, we have identified two non-overlapping interaction domains (ID1 and ID2) within the beta-subunit. ID1 encompasses 118 amino acids at the N-terminus of the mature polypeptide, spanning the cysteine-rich element in this region. ID2, located near the C-terminus, is contained within amino acids 273-400, a region occupied in part by a stretch of acidic residues. Variable usage of 7 alternatively spliced amino acids within ID2 was found not to influence the association of the two subunits. We theorize that the catalytic subunit of glucosidase II binds synergistically to ID1 and ID2, explaining the high associative stability of the enzyme complex.

L43 ANSWER 4 OF 25 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 4
AN 2001:129736 BIOSIS
DN PREV200100129736
TI Active expression of the ubiA gene from E. coli in tobacco: Influence of plant ER-specific signal peptides on the expression of a membrane-bound prenyltransferase in plant cells.
AU Boehm, Robert; Sommer, Susanne; Severin, Klaus; Li, Shu-Ming; Heide, Lutz (1)
CS (1) Pharmazeutische Biologie, Pharmazeutisches Institut, Eberhard-Karls-Universitaet Tuebingen, Auf der Morgenstelle 8, D-72076, Tuebingen: heide@uni-tuebingen.de Germany
SO Transgenic Research, (December, 2000) Vol. 9, No. 6, pp. 477-486. print.
ISSN: 0962-8819.
DT Article
LA English
SL English
AB The ubiA gene from E. coli codes for 4-hydroxybenzoate: polyprenyldiphosphate 3-polyprenyltransferase, an integral membrane protein involved in ubiquinone biosynthesis. This prokaryotic membrane protein was stably expressed in tobacco using Agrobacterium

tumefaciens-mediated transformation. Transgenic lines containing a direct ***fusion*** of the ubiA structural gene to a 35S-derived promoter gave very low enzyme activity levels (average 0.16 pkat/mg). Inclusion of an N-terminal ER-specific signal peptide from a lectin gene from Phaseolus vulgaris resulted in an average activity of 1.08 pkat/mg in the transgenic tobacco lines. The additional inclusion of a C-terminal ***HDEL*** tetrapeptide, responsible for the retention of proteins in the endoplasmic reticulum of eukaryotic cells, increased the activity to 18.6 pkat/mg. When the promotor of this construct was changed from the 35S derivative to the recently described very strong plant promoter (ocs)3mas, the activity increased further to 128.6 pkat/mg. The most active tobacco line showed activities of the introduced enzyme which exceeded those of wild-type E. coli (the source of ubiA) by a factor of 1100. These results demonstrate the efficacy of plant ER-specific signal peptides for the active expression of a prokaryotic membrane protein in plants.

L43 ANSWER 5 OF 25 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 5
AN 2000:129617 BIOSIS
DN PREV200000129617
TI Molecular characterization of a PDI-related gene prpA in Aspergillus niger var. awamori.
AU Wang, Huaming (1); Ward, Michael
CS (1) Genencor International, 925 Page Mill Road, Palo Alto, CA, 94304-1013 USA
SO Current Genetics., (Jan., 2000) Vol. 37, No. 1, pp. 57-64.
ISSN: 0172-8083.
DT Article
LA English
SL English
AB A gene (prpA) homologous to the protein disulfide isomerase gene was isolated from Aspergillus niger by Southern hybridization using the pdiI gene isolated from Trichoderma reesei as a DNA probe. The corresponding cDNA of the prpA gene has also been isolated from an A. niger var. awamori cDNA library. The prpA gene does not belong to any currently recognized family of protein disulfide isomerases since it contains only a single conserved thioredoxin domain at the N-terminus of the protein. The C-terminal two-thirds of the protein has no homology to any known proteins in the database. The PRPA protein contains an ER retention signal (***HDEL***) at its C-terminal end suggesting that it is located in the ER. Southern hybridization at high stringency showed that it was present as a single copy in the genome. Northern hybridization indicated that the transcript level of the prpA gene was higher if the cells were secreting a ***heterologous*** protein, bovine prochymosin. However, over-expression of the prpA gene from a multicopy integrated vector had little effect on chymosin secretion. A strain containing a deletion of the prpA gene was viable. However, deletion of the prpA gene appeared to cause a reduction of bovine chymosin production.

L43 ANSWER 6 OF 25 CAPLUS COPYRIGHT 2001 ACS
AN 2000:559644 CAPLUS
DN 133:131182
TI Insecticidal ***fusion*** protein, its coded gene and method for producing transgenosis strain using said gene
IN Zhu, Zhen; Deng, Chaoyang; Qu, Qiang
PA Genetics Inst., Chinese Academy of Sciences, Peop. Rep. China
SO Faming Zhuanli Shenqing Gongkai Shuomingshu, 55 pp.
CODEN: CNXXEV
DT Patent
LA Chinese
FAN.CNT 1

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI CN 1229087	A	19990922	CN 1999-103430	19990330

AB The disclosed insecticidal ***fusion*** protein contains signal peptide at its N-terminal, insecticidal protein, and endoplasmic reticulum-retention signal at its C-terminal. The signal peptide is selected from potato patatin signal peptide, pathogenesis-related protein PR signal peptide, and soybean Kunitz type trypsin inhibitor (SKTI) signal peptide; the insecticidal protein is selected from Bacillus thuringiensis (Bt) toxoprotein, cowpea trypsin inhibitor (CpTI) insect-resistant protein, paddy mercapto- protease inhibitor (OC), or bivalent insecticidal protein comprising their ***fusion*** proteins; and the signal peptide of the insecticidal protein and endoplasmic reticulum-retention signal such as KDEL and ***HDEL*** . The expression vector is a plant-transfecting vector, contains one or more insecticidal gene expression box and/or other gene expression box, and the exogenous gene of the expression box is controlled under plant promoter. The plant promoter is selected from CaMV 35S promoter, CLCuV replicase gene promoter, paddy actin promoter, T-DNA mas promoter, maize ubiquitin promoter, and their promoter complexes. The expression vector is used for prepn. of insect-resistant plants such as paddy, maize, wheat, tobacco, cotton, soybean, potato, cabbage, brassica oleracea, and pepper, etc. The transgenosis plant is prepd. by construction of expression vector encoding insecticidal ***fusion*** protein, transfecting plant cells with the vector, and culturing the plant cells.

L43 ANSWER 7 OF 25 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 6
AN 1999:444783 BIOSIS
DN PREV199900444783
TI The nuclear envelope serves as an intermediary between the ER and Golgi complex in the intracellular parasite Toxoplasma gondii.
AU Hager, Kristin M.; Striepen, Boris; Tilney, Lewis G.; Roos, David S. (1)
CS (1) Department of Biology, University of Pennsylvania, Philadelphia, PA, 19104-6018 USA
SO Journal of Cell Science, (Aug., 1999) Vol. 112, No. 16, pp. 2631-2638.
ISSN: 0021-9533.
DT Article
LA English
SL English
AB Morphological examination of the highly polarized protozoan parasite Toxoplasma gondii suggests that secretory traffic in this organism progresses from the endoplasmic reticulum to the Golgi apparatus using the nuclear envelope as an intermediate compartment. While the endoplasmic reticulum is predominantly located near the basal end of the parasite, the Golgi is invariably adjacent to the apical end of the nucleus, and the space between the Golgi and nuclear envelope is filled with numerous coatomer-coated vesicles. Staining with antiserum raised against recombinant T. gondii beta-COP confirms its association with the apical juxtanuclear region. Perturbation of protein secretion using brefeldin A, microtubule inhibitors or dithiothreitol disrupts the Golgi, causing swelling of the nuclear envelope, particularly at its basal end. Prolonged drug treatment leads to gross distention of the endoplasmic reticulum, filling the basal end of the parasite. Cloning and sequencing of the T. gondii homolog of the chaperonin protein BiP identifies the carboxy-terminal amino acid sequence ***HDEL*** as this organism's endoplasmic reticulum-retention signal. Appending the ***HDEL*** motif to a recombinant secretory protein (a chimera between the parasite's major surface protein ***fusion*** , P30, and the Green Fluorescent Protein) causes this secretory reporter to be retained intracellularly. P30-GFP-***HDEL*** fluorescence was most intense within the nuclear envelope, particularly at the apical end. These data support a model of secretion in which protein traffic from the endoplasmic reticulum to Golgi occurs via the apical end of the nuclear envelope.

L43 ANSWER 8 OF 25 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 7
AN 1999:483627 BIOSIS
DN PREV199900483627
TI The protein disulphide isomerase gene of the fungus Trichoderma reesei is induced by endoplasmic reticulum stress and regulated by the carbon source.
AU Saloheimo, M. (1); Lund, M.; Penttila, M. E.
CS (1) VTT Biotechnology and Food Research, FIN-02044 VTT, Espoo Finland
SO Molecular and General Genetics, (Aug., 1999) Vol. 262, No. 1, pp. 35-45.
ISSN: 0026-8925.
DT Article
LA English
SL English
AB The gene pdi1 encoding protein disulphide isomerase was isolated from the filamentous fungus Trichoderma reesei by degenerate PCR based on a consensus PDI active-site sequence. It was shown that the Trichoderma pdi1 cDNA is able to complement a yeast mutant with a disrupted PDI1 gene. The putative T. reesei PD1I protein has a predicted 20-amino acid N-terminal signal sequence and the C-terminal fungal consensus ER retention signal ***HDEL*** . The mature protein shows strong conservation relative to other fungal protein disulphide isomerases. The T. reesei pdi1 promoter has two possible unfolded protein response (UPR) elements and it was shown by treatments with dithiothreitol and tunicamycin that the gene is under the control of the UPR pathway. Expression of a ***heterologous*** protein, an IgG antibody Fab fragment, in Trichoderma increases pdi1 expression, probably by inducing the UPR. The level of T. reesei pdi1 mRNA is also regulated by the carbon source, being lowest in glucose-containing media and highest on carbon sources that induce the genes encoding extracellular enzymes. The mechanism of this regulation was studied by examining pdi1 mRNA levels under conditions where the extracellular enzymes are induced by sophorose, as well as in the strain RutC-30, which is mutant for the glucose repressor gene cre1. The results suggest that neither sophorose induction nor glucose repression by the CREI protein affect the pdi1 promoter directly.

L43 ANSWER 9 OF 25 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 8
AN 1998:490652 BIOSIS
DN PREV199800490652
TI Production of human compatible high mannose-type (Man5GlcNAc2) sugar chains in Saccharomyces cerevisiae.
AU Chiba, Yasunori; Suzuki, Misa; Yoshida, Satoshi; Yoshida, Aruto; Ikenaga, Hiroshi; Takeuchi, Makoto (1); Jigami, Yoshifumi; Ichishima, Eiji
CS (1) Central Laboratories Key Technol., KIRIN Brewery Co. Ltd., 1-13-5 Fukuura, Kanazawa-ku, Yokohama 236-0004 Japan
SO Journal of Biological Chemistry, (Oct. 9, 1998) Vol. 273, No. 41, pp. 26298-26304.
ISSN: 0021-9258.
DT Article
LA English
AB A yeast mutant capable of producing Man5GlcNAc2 human compatible sugar chains on glycoproteins was constructed. An expression vector for alpha-1,2-mannosidase with the " ***HDEL*** " endoplasmic reticulum retention/ retrieval tag was designed and expressed in Saccharomyces cerevisiae. An in vitro alpha-1,2-mannosidase assay and Western blot analysis showed that it was successfully localized in the endoplasmic reticulum. A triple mutant yeast lacking three glycosyltransferase activities was then transformed with an alpha-1,2-mannosidase expression vector. The oligosaccharide structures of carboxypeptidase Y as well as cell surface glycoproteins were analyzed, and the recombinant yeast was shown to produce a series of high mannose-type sugar chains including Man5GlcNAc2. This is the first report of a recombinant S. cerevisiae able to produce Man5GlcNAC2-oligosaccharides, the intermediate for

hybrid -type and complex-type sugar chains.

L43 ANSWER 10 OF 25 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 9
AN 1997:163727 BIOSIS
DN PREV199799462930
TI The C-terminal ***HDEL*** sequence is sufficient for retention of secretory proteins in the endoplasmic reticulum (ER) but promotes vacuolar targeting of proteins that escape the ER.
AU Gomord, Veronique; Denmat, Lise-Anne; Fitchette-Laine, Anne-Catherine; Satiat-Jeunemaitre, Beatrice; Hawes, Chris; Faye, Loic (1)
CS (1) LTI-CNRS URA 203, UFR Sci., IFRMP 23, Univ. Rouen, 76821 Mt. St. Aignan Cedex France
SO Plant Journal, (1997) Vol. 11, No. 2, pp. 313-325.
ISSN: 0960-7412.
DT Article
LA English
AB Proteins are co-translationally transferred into the endoplasmic reticulum (ER) and then either retained or transported to different intracellular compartments or to the extracellular space. Various molecular signals necessary for retention in the ER or targeting to different compartments have been identified. In particular, the ***HDEL*** and KDEL signals used for retention of proteins in yeast an animal ER have also been described at the C-terminal end of soluble ER processing enzymes in plants. The ***fusion*** of a KDEL extension to vacuolar proteins is sufficient for their retention in the ER of transgenic plant cells. However, recent results obtained using the same strategy indicate that ***HDEL*** does not contain sufficient information for full retention of phaseolin expressed in tobacco. In the present study, an ***HDEL*** C-terminal extension was fused to the vacuolar or extracellular (DELTA-pro) forms of sporamin. The resulting SpoHDEL or DELTA-proHDEL, as
well as Spo and DELTA-pro, were expressed at high levels in transgenic tobacco cells (Nicotiana tabacum cv BY2). The intracellular location of these different forms of recombinant sporamin was studied by subcellular fractionation. The results clearly indicate that addition of an ***HDEL*** extension to either Spo or DELTA-pro induces accumulation of these sporamin forms in a compartment that co-purifies with the ER markers NADH cytochrome C reductase, binding protein (BiP) and calnexin. In addition, a significant SpoHDEL or DELTA-proHDEL fraction that escapes the ER retention machinery is transported to the vacuole. From these results, it may be proposed that, in addition to its function as an ER retention signal, ***HDEL*** could also act in quality control by targeting chaperones or chaperone-bound proteins that escape the ER to the plant lysosomal compartment for degradation.

L43 ANSWER 11 OF 25 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 10
AN 1997:180572 BIOSIS
DN PREV199799472285
TI Isolation and characterisation of a gene encoding protein disulphide isomerase, pdiA, from Aspergillus niger.
AU Ngiam, C.; Jeenes, D. J. (1); Archer, D. B.
CS (1) Genetics Microbiol. Dep., Inst. Food Res., Norwich Research Park, Colney, Norwich NR4 7UA UK
SO Current Genetics, (1997) Vol. 31, No. 2, pp. 133-138.
ISSN: 0172-8083.
DT Article
LA English
AB Current strategies to improve the secretion of ***heterologous*** proteins from Aspergillus niger include the manipulation of chaperones and foldases specific to the endoplasmic reticulum (ER). Here we report the isolation of a gene, pdiA, encoding a putative protein disulphide isomerase (PDI) from A. niger using the Saccharomyces cerevisiae PDI gene as a probe. Sequencing of a genomic clone and RT-PCR products predict a 515-aa protein comprising a 20-aa ER-translocation signal sequence and a 495-aa mature protein (M-r = 54.3 kDa). The predicted protein also contains two thiol oxidoreductase active sites with a -CGHC- motif and a carboxy terminal - ***HDEL*** ER-retention signal. Three introns were identified within the pdiA gene and Southern- and dot-blot analysis indicates that the gene is present in a single copy. Northern-blot analysis shows a transcript of the predicted size. Sequence homology to a motif associated with protein trafficking and the induction of chaperones has been identified in the pdiA promoter. Transcription of pdiA is induced 3-4-fold after treatment with tunicamycin, an inhibitor of N-linked glycosylation. The kinetics of induction suggest that pdiA expression is not part of the primary stress response.

L43 ANSWER 12 OF 25 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 11
AN 1996:243434 BIOSIS
DN PREV199698791563
TI SNARE-mediated retrograde traffic from the Golgi complex to the endoplasmic reticulum.
AU Lewis, Michael J.; Pelham, Hugh R. B.
CS MRC Lab. Mol. Biol., Hills Rd., Cambridge CB2 2QH UK
SO Cell, (1996) Vol. 85, No. 2, pp. 205-215.
ISSN: 0092-8674.
DT Article
LA English
AB Operation of the secretory pathway in eukaryotic cells requires the selective docking and ***fusion*** of transport vesicles with the appropriate target organelle. This is mediated in part by integral membrane proteins termed v-SNAREs (on vesicles) and t-SNAREs (on the target membranes). We describe a novel yeast t-SNARE that resides on the endoplasmic reticulum and mediates retrograde traffic from the Golgi complex. Mutation of this protein prevents both the ***HDEL*** receptor and a membrane protein bearing a dibasic retrieval signal from recycling to the endoplasmic reticulum. Forward traffic is also blocked, but only indirectly. Comparison with other yeast mutants indicates that Sec21p (gamma-COP) and Sec20p (an endoplasmic reticulum membrane protein)
are also involved primarily, if not exclusively, in retrograde transport.

L43 ANSWER 13 OF 25 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 12
AN 1995:484116 BIOSIS
DN PREV199598498416
TI Production of rat protein disulfide isomerase in Saccharomyces cerevisiae.
AU Laboissiere, Martha C. A.; Chivers, Peter T.; Raines, Ronald T. (1)
CS (1) Dep. Biochem., Univ. Wisconsin-Madison, 420 Henry Mall, Madison, WI 53706-1569 USA
SO Protein Expression and Purification, (1995) Vol. 6, No. 5, pp. 700-706.
ISSN: 1046-5928.
DT Article
LA English
AB Protein disulfide isomerase (PDI) is an abundant protein of the endoplasmic reticulum that catalyzes the oxidation of protein sulfhydryl groups and the isomerization and reduction of protein disulfide bonds. Saccharomyces cerevisiae cells lacking PDI are inviable. PDI is a component of many different protein processing complexes, and the actual activity of PDI that is required for cell viability is unclear. A cDNA that codes for rat PDI fused to the alpha-factor pre-pro segment was expressed in a protease-deficient strain of S. cerevisiae under the control of an ADH2-GAPDH ***hybrid*** promoter. The cells processed the resulting protein and secreted it into the medium as a monomer, despite having a KDEL or ***HDEL*** sequence at its C-terminus. The typical yield of isolated protein was 2 mg per liter of culture. The catalytic activity of the PDI from S. cerevisiae was indistinguishable from that of PDI isolated from bovine liver. This expression system is unique in allowing the same plasmid to be used both to complement pdi1-DELTA S. cerevisiae and to produce PDI for detailed in vitro analyses. Correlations of the in vivo behavior and in vitro properties of PDI are likely to reveal structure-function relationships of biological importance.

L43 ANSWER 14 OF 25 CAPLUS COPYRIGHT 2001 ACS
AN 1995:691522 CAPLUS
DN 123:280986
TI Degradation of transport-competent destabilized phaseolin with a signal for retention in the endoplasmic reticulum occurs in the vacuole
AU Pueyo, Jose J.; Chrispeels, Maarten J.; Herman, Eliot M.
CS Dep. Biol., Univ. California, San Diego, La Jolla, CA, 92093-0116, USA
SO Planta (1995), 196(3), 586-96
CODEN: PLANAB; ISSN: 0032-0935
DT Journal
LA English
AB To understand how plant cells exert quality control over the proteins that pass through the secretory system we examd. the transport and accumulation of the bean (Phaseolus vulgaris L.) vacuolar storage protein phaseolin, structurally modified to contain a helix-breaking epitope and carboxyterminal ***HDEL*** , an endoplasmic reticulum (ER)-retention signal. The constructs were expressed in tobacco (Nicotiana tabacum L.) with a seed-specific promoter. The results show that phaseolin0HDEL accumulates in the protein-storage vacuoles, indicating that HEDL does not contain sufficient information for retention in the ER. However, the ER of seeds expressing the phaseolin- ***HDEL*** construct contain relatively more phaseolin- ***HDEL*** compared to phaseolin in the ER of seeds expressing the phaseolin construct. This result indicates that the flow out of the ER is retarded but not arrested by the presence of ***HDEL*** . Introduction into phaseolin of the epitope "himet" greatly reduces the accumulation of HiMet phaseolin compared to normal phaseolin. However, the increased abundance within the ER is similar for both phaseolin- ***HDEL*** and HiMet phaseolin- ***HDEL*** . Using immunocytochem. with specific antibodies, HiMet phaseolin was found in the ER, the Golgi stack, and in transport vesicles indicating that it was transport competent. It was also present at an early stage of seed development in the protein-storage vacuoles, but was not found there at later stages of seed development. Together these results support the conclusion that the HiMet epitope did not alter the structure of the protein sufficiently to make it transport incompetent. However, the protein was sufficiently destabilized to be degraded by vacuolar proteases.

L43 ANSWER 15 OF 25 CAPLUS COPYRIGHT 2001 ACS
AN 1995:71865 CAPLUS
DN 122:25001
TI Molecular cloning of a fungal cDNA encoding protein disulfide isomerase
AU Kajino, Tsutomu; Sarai, Kiyoko; Imaeda, Takao; Idekoba, Chie; Asami, Osamu; Yamada, Yukio; Hirai, Masana; Udaka, Shigezo
CS Toyota Cent. Res. Dev. Lab. Inc., Aichi, 480-11, Japan
SO Biosci., Biotechnol., Biochem. (1994), 58(8), 1424-9
CODEN: BBBIEJ; ISSN: 0916-8451
DT Journal
LA English
AB Based on the partial amino acid sequences of a protein disulfide isomerase (PDI) from Humicola insolens, two primers were synthesized for reverse transcriptase mediated polymerase chain reaction (RT-PCR) of a fungal RNA. A 0.2-kbp fragment around the consensus sequence of PDIs was obtained and used as a probe for screening a fungal cDNA library. A cDNA clone of PDI

from H. insolens was isolated and encoded a polypeptide consisting of 505 amino acids, which was characterized by a N-terminal signal sequence composed of 20 amino acids, a consensus sequence (WCGHCK) at two positions, and a C-terminal endoplasmic reticulum retention signal (***HDEL***). Bacillus brevis harboring an expression plasmid bearing the fungal PDI cDNA was prepd. and its culture supernatant showed a significant PDI activity. This indicates that glycosylation of a fungal PDI is not essential for the enzymic activity related to an interchange of disulfide bonds.

L43 ANSWER 16 OF 25 CAPLUS COPYRIGHT 2001 ACS
AN 1994:98497 CAPLUS
DN 120:98497
TI Method for increasing production of disulfide bonded recombinant proteins by (saccharomyces cerevisiae)
IN Tuite, Michael F.; Freedman, Robert B.; Schultz, Loren D.; Ellis, Ronald W.; Markus, Henry Z.; Montgomery, Donna L.
PA Merck and Co., Inc., USA; University of Kent
SO PCT Int. Appl., 107 pp.
CODEN: PIXXD2
DT Patent
LA English
FAN.CNT 1

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI WO 9325676	A1	19931223	WO 1993-US5318	19930602
W: AU, BB, BG, BR, CA, CZ, FI, HU, JP, KR, KZ, LK, MG, MN, MW, NO, NZ, PL, RO, RU, SD, SK, UA, US				
RW: AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG				
AU 9345274	A1	19940104	AU 1993-45274	19930602
AU 679448	B2	19970703		
JP 07508881	T2	19951005	JP 1993-501587	19930602
EP 746611	A1	19961211	EP 1993-915201	19930602
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LI, LU, NL, PT, SE				
PRAI US 1992-901713		19920612		
WO 1993-US5318		19930602		

AB A method for increasing the yield of disulfide-bonded proteins produced by expression of the gene in yeast, esp. secreted proteins is described. The method uses host strains of Saccharomyces cerevisiae showing regulated overprodn. yeast or human protein disulfide isomerase (PDI) that catalyzes the formation of disulfide bonds in secretory and cell-surface proteins. These strains show greatly increased secretion of disulfide-bonded proteins of potential therapeutic significance. These strains have the potential to increase the yields of various disulfide-bonded proteins. The yeast gene (PDI1) was cloned by screening a Sau3A partial digest library in pMA3a by screening with an oligonucleotide corresponding to the conserved thioredoxin-like active site. A host strain with the PDI1 gene inactivated by insertion of the HIS3 gene was constructed and the gene was placed under control of the LYS2 or URA3 loci. The corresponding human was also introduced into yeast and a series of measures including selection of the signal sequence and improvement of the membrane anchor were used to increase activity of the enzyme. A gene for the disulfide bond-rich blood-coagulation factor Xa inhibitor antistasin was cloned from Haementeria officinalis and introduced into a no. of these strains using the expression vector pKH4.alpha.2. Yields of activity from yeasts carrying these PDI expression cassettes were increased up to 3-fold over control strains.

L43 ANSWER 17 OF 25 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 13
AN 1993:164437 BIOSIS
DN PREV199395085487
TI Cloning of the gene encoding a Schistosoma mansoni antigen homologous to human Ro/SS-A autoantigen.
AU Khalife, Jamal (1); Trottein, Francois; Schacht, Anne-Marie; Godin, Claude; Pierce, Raymond J.; Capron, Andre
CS (1) Centre d'Immunologie et de Biologie Parasitaire, Unite Mixte INSERM U167-CNRS URA 624, Institut Pasteur de Lille, 1 rue du Prof. Calmette, B.P. 245, 59019 Lille Cedex France
SO Molecular and Biochemical Parasitology, (1993) Vol. 57, No. 2, pp. 193-202.
ISSN: 0166-6851.
DT Article
LA English
AB A cDNA library was constructed from the mRNA of adult worms of Schistosoma mansoni in the expression vector lambda-gt11 and screened with a rabbit antiserum raised against a 60-65 kDa electroeluted adult worm fraction. Two overlapping clones were selected and a partial nucleotide sequence was deduced (1172 bp). The full-length sequence was obtained by the amplification of the 5' end of the first strand cDNA using PCR. The overall mRNA size was 1335 nt including a 25 nt 5' non-coding region and a 131 nt untranslated region with the poly(A) tail. The predicted amino acid sequence of 393 aa (45 kDa) has 52% identity with the human Ro/SS-A autoantigen, which is considered to be the human calreticulin. As for the human Ro/SS-A, the protein encoded by the cDNA described here contains a hydrophobic leader sequence and a carboxyl terminal sequence. ***HDEL*** consensus signal sequence for retention in the ER. An antiserum raised against the ***fusion*** protein of one clone recognized a 58-kDa antigen in homogenates of cercariae and of adult worms. The expression of the protein in the pGEX-2T ***fusion*** system allowed us to show the presence of specific antibodies in S. mansoni infected patients' sera and in the sera of patients with systemic lupus erythematosus, reflecting a cross-immunoreactivity between the S. mansoni protein and the human calreticulin autoantigen.

L43 ANSWER 18 OF 25 EMBASE COPYRIGHT 2001 ELSEVIER SCI. B.V.
AN 92170487 EMBASE
DN 1992170487
TI Plant and mammalian sorting signals for protein retention in the endoplasmic reticulum contain a conserved epitope.
AU Denecke J.; De Rycke R.; Botterman J.
CS University of Agricultural Sciences, Uppsala Genetic Centre, Department of Molecular Genetics, Box 7003,S-75007 Uppsala, Sweden
SO EMBO Journal, (1992) 11/6 (2345-2355).
ISSN: 0261-4189 CODEN: EMJODG
CY United Kingdom
DT Journal; Article
FS 029 Clinical Biochemistry
LA English
SL English
AB We studied protein sorting signals which are responsible for the retention of reticuloplasmins in the lumen of the plant endoplasmic reticulum (ER). A non-specific passenger protein, previously shown to be secreted by default, was used as a carrier for such signals. Tagging with C-terminal tetrapeptide sequences of mammalian (KDEL) and yeast (***HDEL***) reticuloplasmins led to effective accumulation of the protein chimeras in the lumen of the plant ER. Some single amino acid substitutions within the tetrapeptide tag (SDEL, -KDDL, -KDEI and -KDEV) can cause a complete loss of its function as a retention signal, demonstrating the high specificity of the retention machinery. However, other modifications confer efficient (-RDEL) or partial (-KEEL) retention. It is also shown that the efficiency of protein retention is not significantly impaired by an increased ligand concentration in plants. The efficiently retained chimeras (-KDEL, -***HDEL*** and -RDEL) were shown to be recognized by a monoclonal antibody directed against the C-terminus of the mammalian reticuloplasmin protein disulfide isomerase (PDI). The recognized epitope is also present in several putative reticuloplasmins in microsomal fractions of plant and mammalian cells, suggesting that the antibodies recognize an important structural determinant of the retention signal. In addition, data are discussed which support the view that upstream sequences beyond the C-terminal tetrapeptide can influence or may be part of the structure of reticuloplasmin retention signals.

L43 ANSWER 19 OF 25 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 14
AN 1992:280227 BIOSIS
DN BA94:4877
TI ANALYSIS OF THE BIP GENE AND IDENTIFICATION OF AN ER RETENTION SIGNAL IN SCHIZOSACCHAROMYCES-POMBE.
AU PIDOUX A L; ARMSTRONG J
CS MEMBRANE MOL. BIOL. LAB., IMPERIAL CANCER RES. FUND, BOX 123, LINCOLN'S INN FIELDS, LONDON, WC2A 3PX, UK.
SO EMBO (EUR MOL BIOL ORGAN) J, (1992) 11 (4), 1583-1591.
CODEN: EMJODG. ISSN: 0261-4189.
FS BA; OLD
LA English
AB We have cloned the gene for the resident luminal ER protein BiP from the fission yeast, Schizosaccharomyces pombe. The predicted protein product is equally divergent from the budding yeast and mammalian homologues. Disruption of the BiP gene in S. pombe is lethal and BiP mRNA levels are regulated by a variety of stresses including heat shock. Immunofluorescence of cells expressing an epitope-tagged BiP protein show it to be localized to the nuclear envelope, around the cell periphery and in a reticular structure through the cytoplasm. Unexpectedly, we find the BiP protein contains an N-linked glycosylation site which can be utilized. The C-terminal four amino acids of BiP are Ala-Asp-Glu-Leu, a new variant of the XDEL sequence found at the C-termini of luminal endoplasmic reticulum proteins. To determine whether this sequence acts as a sorting signal in S. pombe we expressed an acid phosphatase ***fusion*** protein extended at its C-terminus with the amino acids ADEL. Analysis of the sorting of this ***fusion*** protein indicates that the ADEL sequence is sufficient to cause the retention of proteins in the endoplasmic reticulum. The sequences DDEL, ***HDEL*** and KDEL can also direct ER-retention of acid phosphatase in S. pombe.

L43 ANSWER 20 OF 25 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 15
AN 1993:117013 BIOSIS
DN PREV199395061113
TI A mutant Kex2 enzyme with a C-terminal ***HDEL*** sequence releases correctly folded human insulin-like growth factor-1 from a precursor accumulated in the yeast endoplasmic reticulum.
AU Chaudhuri, Bhabatosh (1); Latham, Sarah E.; Stephan, Christine
CS (1) Dep. Biotechnol., K-681.1.06, Ciba-Geigy Ltd., CH-4002 Basel Switzerland
SO European Journal of Biochemistry, (1992) Vol. 210, No. 3, pp. 811-822.
ISSN: 0014-2956.
DT Article
LA English
AB Mutations in the pro region of the yeast DNA ***hybrid*** of prepro-alpha-factor and human insulin-like growth factor-1 (IGF-1) cause the accumulation, in the yeast Saccharomyces cerevisiae, of an unglycosylated precursor protein where the pre sequence is missing. The prepro sequence of the prepro-a-factor consists of a pre or signal sequence and a proregion which possesses three sites for N-glycosylation.

Isolation of a precursor, where the pro region is still linked to IGF-1 through a pair of dibasic amino acid residues, implies that the polypeptide may have translocated into the endoplasmic reticulum (ER) but has not been processed by the Golgi membrane-bound Kex2 endoprotease. However, the lack of any N-glycosylation in the translocated polypeptide is surprising. The mutated pro region, can be processed, in vitro, by treatment with a soluble form of the Kex2 enzyme. It is also possible to release the pro region, in vivo, by coexpressing a mutant Kex2 protease which is partially retained in the ER with the help of the C-terminal tetrapeptide sequence, ***HDEL*** . The mature IGF-1, which is secreted from the intracellular pool of precursor proteins, is predominantly an active, monomeric molecule, corroborating observations that early removal of the pro region before folding in the ER helps to prevent aberrant intermolecular disulfide-bond formation in IGF-1. These results have revealed the utility of the ER-retained Kex2 enzyme as a novel in vivo biochemical tool.

L43 ANSWER 21 OF 25 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 16
AN 1992:209003 BIOSIS
DN BA93:109228
TI A NOVEL KEX2 ENZYME CAN PROCESS THE PROREGION ON THE YEAST ALPHA-FACTOR
LEADER IN THE ENDOPLASMIC RETICULUM INSTEAD OF IN THE GOLGI.
AU CHAUDHURI B; LATHAM S E; HELLIWELL S B; SEEBOTH P
CS DEP. BIOTECHNOLOGY K-681.106, CIBA-GEIGY LTD., CH-4002 BASEL, SWITZERLAND.
SO BIOCHEM BIOPHYS RES COMMUN, (1992) 183 (1), 212-219.
CODEN: BBRCA9. ISSN: 0006-291X.
FS BA; OLD
LA English
AB The prepro sequence of the yeast prepro-.alpha.-factor, usually referred to as the .alpha.-factor leader, has often been used for the efficient secretion of ***heterologous*** proteins from the yeast Saccharomyces cerevisiae. The .alpha.-factor leader consists of a 19-amino acid N-terminal pre or signal sequence followed by a 66-amino acid proregion. After removal of the signal sequence during membrane translocation, the proregion is cleaved from the precursor protein by the Kex2 endoprotease only in a late Golgi compartment. Here we report that a modified Kex2 enzyme, containing at the C-terminus the ***HDEL*** tetrapeptide, cleaves the proregion from the .alpha.-factor leader-human insulin like growth factor-1 ***fusion*** in the endoplasmic reticulum. The processing of pro-proteins earlier in the secretion pathway could be helpful in defining the cellular function of the proregions present naturally in various eucaryotic precursor proteins.

L43 ANSWER 22 OF 25 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 17
AN 1990:218116 BIOSIS
DN BA89:115406
TI ERD1 A YEAST GENE REQUIRED FOR THE RETENTION OF LUMINAL ENDOPLASMIC
RETICULUM PROTEINS AFFECTS GLYCOPROTEIN PROCESSING IN THE GOLGI APPARATUS.
AU HARDWICK K G; LEWIS M J; SEMENZA J; BEAN N; PELHAM H R B
CS MRC LAB. MOLECULAR BIOL., HILLS ROAD, CAMBRIDGE CB2 2QH, UK.
SO EMBO (EUR MOL BIOL ORGAN) J, (1990) 9 (3), 623-630.
CODEN: EMJODG. ISSN: 0261-4189.
FS BA; OLD
LA English
AB We have previously shown that the C-terminal sequence ***HDEL*** acts as a retention signal for luminal endoplasmic reticulum (ER) proteins in Saccharomyces cerevisiae, and that it is possible to isolate mutants that fail to retain an invertase ***fusion*** protein bearing this signal. Analysis of many such mutants defines two genes, ERD1 and ERD2. Cells lacking the ERD1 gene secrete the endogenous ER protein, BiP. Under normal growth conditions, the rate of secretion is equivalent to the rate at which wild-type cells secrete a modified form of BiP that lacks the ***HDEL*** signal altogether. Thus, erd1 cells show a profound disruption of the retention system. The mutant cells have no gross abnormality of their intracellular membrane system, but show defects in the Golgi-dependent modification of glycoproteins. We suggest that sorting of luminal ER proteins normally occurs in the Golgi, and that the function of ERD1 is required for the correct interaction of an ***HDEL*** receptor with its ligands. The sequence of ERD1 predicts a membrane protein with several transmembrane domains, a conclusion supported by analysis of ERD1-SUC2 ***fusion*** proteins.

L43 ANSWER 23 OF 25 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 18
AN 1990:430894 BIOSIS
DN BA90:91695
TI RECYCLING OF PROTEINS FROM THE GOLGI COMPARTMENT TO THE ER IN YEAST.
AU DEAN N; PELHAM H R B
CS MEDICAL RESEARCH COUNCIL LABORATORY MOLECULAR BIOLOGY, CAMBRIDGE CB2 2QH,
U.K.
SO J CELL BIOL, (1990) 111 (2), 369-378.
CODEN: JCLBA3. ISSN: 0021-9525.
FS BA; OLD
LA English
AB In the yeast Saccharomyces cerevisiae, the carboxyl terminal sequence His-Asp-Glu-Leu (***HDEL***) has been shown to function as an ER retention sequence (Pelham, H. R. B., K. G. Hardwick, and M. J. Lewis. 1988. EMBO (Eur. Mol. Biol. Organ.) J. 7:1757-1762). To examine the mechanism of retention of soluble ER proteins in yeast, we have analyzed the expression of a preproalpha factor ***fusion*** protein, tagged at the carboxyl terminus with the ***HDEL*** sequence. We demonstrate that this ***fusion*** protein, expressed in vivo, accumulates intracellularly as a precursor contaiing both ER and Golgi-specific oligosaccharide modifications. The Golgi-specific carbohydrate modification, which occurs in a SEC18-dependent manner consists of .alpha.1-6 mannose linkages, with no detectable .alpha.1-3 mannose additions, indicating that the transit of the ***HDEL*** -tagged ***fusion*** protein is confined to an early Golgi compartment. Results obtained from the fractionation of subcellular organelles from yeast expressing ***HDEL*** -tagged ***fusion*** proteins suggest that the Golgi-modified species are present in the ER. Overexpression of ***HDEL*** -tagged preproalpha factor results in the secretion of an endogenous ***HDEL*** -containing protein, demonstrating that the ***HDEL*** recognition system can be saturated. These results support the model in which the retention of these proteins in the ER is dependent on their receptor-mediated recycling from the Golgi complex back to the ER.

L43 ANSWER 24 OF 25 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 19
AN 1989:404377 BIOSIS
DN BA88:73802
TI KAR2 A KARYOGAMY GENE IS THE YEAST HOMOLOG OF THE MAMMALIAN BIP-GRP78
GENE.
AU ROSE M D; MISRA L M; VOGEL J P
CS DEP. BIOL., LEWIS THOMAS LAB., PRINCETON UNIV., PRINCETON, NEW JERSEY
08544-1014.
SO CELL, (1989) 57 (7), 1211-1222.
CODEN: CELLB5. ISSN: 0092-8674.
FS BA; OLD
LA English
AB The yeast KAR2 gene was isolated by complementation of a mutation that blocks nuclear ***fusion*** . The predicted KAR2 protein sequence is most homologous to mammalian BiP/GRP78 and has several structural features in common with it: a functional secretory signal sequence, a yeast endoplasmic reticulum retention signal (***HDEL***) at the carboxyl terminus, and the absence of potential N-linked glycosylation sites. Moreover KAR2 is regulated like BiP/GRP78: the level of mRNA is increased by drug treatments and mutations that cause accumulation of secretory precursors in the endoplasmic reticulum. However, unlike BiP/GRP78, KAR2 is also regulated by heat shock. Deletion of the KAR2 gene generated a recessive lethal mutation, showing that BiP/GRP78 function is required for cell viability.

L43 ANSWER 25 OF 25 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 20
AN 1989:94018 BIOSIS
DN BA87:48154
TI SORTING OF SOLUBLE ER PROTEINS IN YEAST.
AU PELHAM H R B; HARDWICK K G; LEWIS M J
CS MRC LAB. MOL. BIOL., HILLS ROAD, CAMBRIDGE CB2 2QH, UK.
SO EMBO (EUR MOL BIOL ORGAN) J, (1988) 7 (6), 1757-1762.
CODEN: EMJODG. ISSN: 0261-4189.
FS BA; OLD
LA English
AB In animal cells, luminal endoplasmic reticulum (ER) proteins are prevented from being secreted by a sorting system that recognizes the C-terminal sequence KDEL. We show that yeast has a similar sorting system, but it recognizes ***HDEL*** , rather than KDEL: derivatives of the enzyme invertase that bear the ***HDEL*** signal fail to be secreted. An invertase ***fusion*** protein that is retained in the cells is partially modified by outer-chain mannosyl transferase, which reside in the Golgi element. This supports the view, based on studies in animal cells, that ER targeting is achieved by continuous retrieval of proteins from the Golgi. We have used an invertase ***fusion*** gene to screen for mutants that are defective in this sorting system. Over 60 mutants were obtained; eight of these are alleles of a single gene, erd1. The mutant strains grow normally at 30.degree. C, but instead of retaining the ***fusion*** protein in the cells, they secrete it.

=> d bib abs l41 1-
YOU HAVE REQUESTED DATA FROM 92 ANSWERS - CONTINUE? Y/(N):y

L41 ANSWER 1 OF 92 CAPLUS COPYRIGHT 2001 ACS
AN 2001:45049 CAPLUS
DN 134:97534
TI Conjugates for the delivery of active substances into cells, cell compartments and membranes
IN Braun, Klaus; Friedrich, Eckart; Waldeck, Waldemar; Peschke, Peter; Pipkorn, Ruediger; Debus, Juergen
PA Deutsches Krebsforschungszentrum Stiftung des Oeffentlichen Rechts, Germany
SO Ger. Offen., 10 pp.
CODEN: GWXXBX
DT Patent
LA German
FAN.CNT 1

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI DE 19933492	A1	20010118	DE 1999-19933492	19990716

WO 2001005432 A2 20010125 WO 2000-DE2346 20000714
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CR, CU, CZ, DK, DM, DZ, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG
PRAI DE 1999-19933492 A 19990716
AB The invention concerns the prodn. and application of conjugates for the delivery of active substance into cells, cell compartments and membranes that contain fragments of a penetrating protein, a target-specific localization protein and the active substance. Cell-penetrating proteins are penetratin, transportan or their derivs. Sequences of the target-specific localization peptides are given for endoplasmic reticulum, mitochondria, nucleus, peroxisomes and cell membrane. Active substances are diagnostic agents or drugs. Spacers can be included into the conjugate between the active substance and the target-specific peptide. Synthesis methods include the Merrifield synthesis and coupling of the non-peptide component. Thus penetratin, a nuclear localization sequence and a spacer sequence peptide-conjugate was synthesized; after purifn., it was coupled with rhodamine 110. The conjugate was incubated with AT-1 and DU-145 cells; the penetration of the rhodamine 110 contg. conjugate into the nucleus was detected by fluoroscence microscopy.
RE.CNT 3
RE
(1) Anon; Drug Design 1980, VX, PS226
(2) Anon; Molecular Biology of the Cell 1983, PS344
(3) Anon; Rompp Chemie Lexikon 1998, V10, Ps2584

L41 ANSWER 2 OF 92 CAPLUS COPYRIGHT 2001 ACS
AN 2001:549120 CAPLUS
TI Ykt6 forms a SNARE complex with syntaxin 5, GS28, and Bet1 and participates in a late stage in endoplasmic reticulum-Golgi transport
AU Zhang, Tao; Hong, Wanjin
CS Membrane Biology Laboratory, Institute of Molecular and Cell Biology, Singapore, 117609, Singapore
SO J. Biol. Chem. (2001), 276(29), 27480-27487
CODEN: JBCHA3; ISSN: 0021-9258
PB American Society for Biochemistry and Molecular Biology
DT Journal
LA English
AB The yeast SNARE Ykt6p has been implicated in several trafficking steps, including vesicular transport from the endoplasmic reticulum (ER) to the Golgi, intra-Golgi transport, and homotypic vacuole ***fusion*** . The functional role of its mammalian homolog (Ykt6) has not been established. Using antibodies specific for mammalian Ykt6, it is revealed that it is found mainly in Golgi-enriched membranes. Three SNAREs, syntaxin 5, GS28, and Bet1, are specifically assocd. with Ykt6 as revealed by co-immunopptn., suggesting that these four SNAREs form a SNARE complex. Double labeling of Ykt6 and the Golgi marker mannosidase II or the ER-Golgi recycling marker ***KDEL*** receptor suggests that Ykt6 is primarily assocd. with the Golgi app. Unlike the ***KDEL*** receptor, Ykt6 does not cycle back to the peripheral ER exit sites. Antibodies against Ykt6 inhibit in vitro ER-Golgi transport of vesicular stomatitis virus envelope glycoprotein (VSVG) only when they are added before the EGTA-sensitive stage. ER-Golgi transport of VSVG in vitro is also inhibited by recombinant Ykt6. In the presence of antibodies against Ykt6, VSVG accumulates in peri-Golgi vesicular structures and is prevented from entering the mannosidase II compartment, suggesting that Ykt6 functions at a late stage in ER-Golgi transport. Golgi app. marked by mannosidase II is fragmented into vesicular structures in cells microinjected with Ykt6 antibodies. It is concluded that Ykt6 functions in a late step of ER-Golgi transport, and this role may be important for the integrity of the Golgi complex.
RE.CNT 64
RE
(1) Aridor, M; J Cell Biol 1995, V131, P875 CAPLUS
(3) Banfield, D; J Cell Biol 1994, V127, P357 CAPLUS
(4) Beckers, C; Cell 1987, V50, P523 CAPLUS
(5) Bock, J; J Biol Chem 1996, V271, P17961 CAPLUS
(6) Bock, J; Nature 2001, V409, P839 CAPLUS
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L41 ANSWER 3 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 1
AN 2001:300311 BIOSIS
DN PREV200100300311
TI Isolation of new anti-CD30 scFvs from DNA-immunized mice by phage display and biologic activity of recombinant immunotoxins produced by ***fusion*** with truncated Pseudomonas exotoxin.
AU Rozemuller, Hendrick; Chowdhury, Partha S.; Pastan, Ira; Kreitman, Robert J. (1)
CS (1) Laboratory of Molecular Biology, National Cancer Institute, National Institutes of Health, 37 Convent Drive, 37/4B27, Bethesda, MD: kreitmar@mail.nih.gov USA
SO International Journal of Cancer, (15 June, 2001) Vol. 92, No. 6, pp. 861-870. print.
ISSN: 0020-7136.
DT Article
LA English
SL English
AB To target CD30 on Hodgkin's disease and anaplastic large-cell lymphoma, anti-CD30 single-chain antibodies were obtained by DNA immunization of mice with the complete human CD30 cDNA. Spleens were isolated from mice with high anti-CD30 titer, and the RNA was used for the production of an scFv-displaying phage library. Specific phages were enriched by 3 rounds of panning on soluble CD30 or CD30+ K562 cells. Recombinant immunotoxins (rITs) were made from 3 ELISA-positive scFv phages by ***fusion*** to a 38 kDa truncated mutant of Pseudomonas exotoxin (PE38) with or without a ***KDEL*** mutant sequence at the C terminus. In vitro cytotoxicity of purified anti-CD30 rITs was measured on CD30-transfected A431 cells. IC50 values ranged from 3 to 7 ng/ml (50-110 pM) for PE38 rITs and 0.1 ng/ml (2 pM) for the PE38- ***KDEL*** IT on A431-CD30 cells. The parental A431 cells were resistant, indicating that the cytotoxicity was specific and CD30-mediated. rITs were tested for anti-tumor activity in a nude mouse model. A431-CD30 cells were injected s.c. on day 0; then, mice bearing measurable tumors were treated beginning on day 4 with 3 alternate daily doses i.v. Anti-tumor activity was dose-dependent and not found when irrelevant ITs were administered or when CD30- tumors were treated. Our data show that DNA immunization and antibody phage display may be useful in producing new rITs against hematologic malignancies.

L41 ANSWER 4 OF 92 EMBASE COPYRIGHT 2001 ELSEVIER SCI. B.V.DUPLICATE 2
AN 2001294929 EMBASE
TI Intracellular apolipoprotein E affects Amyloid Precursor Protein processing and amyloid A.beta. production in COS-1 cells.
AU Hass S.; Weidemann A.; Utermann G.; Baier G.
CS G. Baier, Inst. for Med. Biol./Human Genetics, University of Innsbruck, Schoepfstr. 41, A-6020 Innsbruck, Austria. Gottfried.Baier@uibk.ac.at
SO Molecular Genetics and Genomics, (2001) 265/5 (791-800).
Refs: 74
ISSN: 1617-4615 CODEN: MGGOAA
CY Germany
DT Journal; Article
FS 029 Clinical Biochemistry
LA English
SL English
AB The apoE gene has been identified as a major susceptibility locus for late-onset Alzheimer's disease (LOAD) The .epsilon.4 allele greatly reduces age of onset of LOAD as compared to the wild-type .epsilon.3 allele. The molecular mechanism(s) underlying the association has not yet been fully elucidated. The apoE protein has been shown to physically interact with the A.beta. region of the Amyloid Precursor Protein (APP), but also with the ectodomain of the APP holoprotein itself. In this study we have used apoE ***fusion*** proteins containing either the ER retention sequence ***KDEL*** or trans-Golgi network (TGN) signal sequence in order to define potential apoE-mediated alterations in APP protein processing. Co-expression and pulse-chase experiments showed that a functional apoE:APP interaction occurs intracellularly which directly affects maturation and subsequently the secretion kinetics of APP. In addition, an .epsilon.4 allele-specific induction of A.beta. production has been demonstrated, apoE3 resulted in increased A.beta. production only when targeted to the ER, as observed in cells transfected with an apoE3KDEL ***fusion*** protein as well as following treatment with brefeldin A. The findings suggest that in cells that express both apoE and APP, such as astrocytes and microglia, a functional apoE:APP interaction may occur which modulates APP processing and A.beta. production.

L41 ANSWER 5 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 3
AN 2001:257699 BIOSIS
DN PREV200100257699
TI Expression of a sulphur-rich sunflower albumin gene in transgenic tall fescue (Festuca arundinacea Schreb.) plants.
AU Wang, Z. Y.; Ye, X. D.; Nagel, J.; Potrykus, I.; Spangenberg, G. (1)
CS (1) Plant Biotechnology Centre, Agriculture Victoria and CRC for Molecular Plant Breeding, La Trobe University, Bundoora, Victoria, 3083: German.Spangenberg@nre.vic.gov.au Australia
SO Plant Cell Reports, (March, 2001) Vol. 20, No. 3, pp. 213-219. print.
ISSN: 0721-7714.
DT Article
LA English
SL English
AB Transgenic tall fescue (Festuca arundinacea Schreb.) plants have been generated that express foreign genes encoding a rumen-stable protein rich in sulphur-containing amino acids. The aim was to improve the protein quality of a forage grass for ruminant nutrition. ***Chimeric*** sulphur-rich sunflower albumin (SFA8) genes, including an endoplasmic reticulum retention signal (***KDEL***), were constructed under the control of constitutive (CaMV 35S) and light-regulated (wheat Cab) promoters. These constructs were introduced into the tall fescue genome by microprojectile bombardment of embryogenic suspension cells. The sunflower albumin transgenes stably integrated into the plant genome as demonstrated by Southern hybridization analysis. The transgenic tall fescue plants produced the expected transcript, and the corresponding sulphur-rich SFA8 protein accumulated up to 0.2% of the total soluble protein in individual transgenic plants.

L41 ANSWER 6 OF 92 CAPLUS COPYRIGHT 2001 ACS
AN 2001:532883 CAPLUS
TI ***KDEL*** -cargo regulates interactions between proteins involved in COPI vesicle traffic: measurements in living cells using FRET
AU Majoul, Irina; Straub, Martin; Hell, Stefan W.; Duden, Rainer; Soling, Hans-Dieter

CS Department of Neurobiology, Max-Planck-Institute of Biophysical Chemistry, Gottingen, D-37077, Germany
SO Dev. Cell (2001), 1(1), 139-153
CODEN: DCEEBE; ISSN: 1534-5807
PB Cell Press
DT Journal
LA English
AB How the occupied ***KDEL*** receptor ERD2 is sorted into COPI vesicles for Golgi-to-ER transport is largely unknown. Here, interactions between proteins of the COPI transport machinery occurring during a "wave" of transport of a ***KDEL*** ligand were studied in living cells. FRET between CFP and YFP ***fusion*** proteins was measured by multifocal multiphoton microscopy and bulk-cell spectrofluorimetry. Ligand binding induces oligomerization of ERD2 and recruitment of ARFGAP to the Golgi, where the (ERD2)n/ARFGAP complex interacts with membrane-bound ARF1. During ***KDEL*** ligand transport, interactions of ERD2 with .beta.-COP and p23 decrease and the proteins segregate. Both p24a and p23 interact with ARF1, but only p24 interacts with ARFGAP. These findings suggest a model for how cargo-induced oligomerization of ERD2 regulates its sorting into COPI-coated buds.
RE.CNT 51
RE
(1) Aoe, T; EMBO J 1997, V16, P7305 CAPLUS
(2) Aoe, T; Proc Natl Acad Sci USA 1998, V95, P1624 CAPLUS
(3) Barlowe, C; Traffic 2000, V1, P371 CAPLUS
(5) Blum, R; J Cell Sci 1999, V112, P537 CAPLUS
(6) Bremser, M; Cell 1999, V96, P495 CAPLUS
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L41 ANSWER 7 OF 92 CAPLUS COPYRIGHT 2001 ACS
AN 2000:368420 CAPLUS
DN 133:3725
TI Suppression of xenotransplant rejection
IN Ramrakha, Punit Satyavrat; George, Andrew John Timothy; Haskard, Dorian; Lechler, Robert Ian
PA Imperial College Innovations Limited, UK
SO PCT Int. Appl., 36 pp.
CODEN: PIXXD2
DT Patent
LA English
FAN.CNT 1

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI WO 2000031126	A2	20000602	WO 1999-GB3888	19991122
WO 2000031126	A3	20000824		

W: AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CR, CU, CZ, DE, DK, DM, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM
RW: GH, GM, KE, LS, MW, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG

EP 1131411	A2	20010912	EP 1999-956179	19991122

R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO

PRAI GB 1998-25555 A 19981120
WO 1999-GB3888 W 19991122
AB The authors disclose methods for suppression of graft rejection, particularly xenograft rejection. In one example, a phage library was created for human antibodies directed to porcine VCAM. Phage-derived scFvs were engineered to express the endoplasmic reticulum targeting signal ***KDEL*** and transfected into porcine aortic endothelial cells. FACS anal. showed a redn. in VCAM surface expression and a functional loss in adhesive function as demonstrated by reduced binding to Jurkat cells.

L41 ANSWER 8 OF 92 CAPLUS COPYRIGHT 2001 ACS
AN 2000:98760 CAPLUS
DN 132:133894
TI Inhibition of ***KDEL*** receptor-mediated return of heat shock protein complexes to the endoplasmic reticulum and their adjuvant use
IN Rothman, James E.; Mayhew, Mark; Hoe, Mee H.
PA Sloan-Kettering Institute for Cancer Research, USA
SO PCT Int. Appl., 87 pp.
CODEN: PIXXD2
DT Patent
LA English
FAN.CNT 1

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI WO 2000006729	A1	20000210	WO 1999-US17147	19990728

W: AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM
RW: GH, GM, KE, LS, MW, SD, SL, SZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG

US 6160088	A	20001212	US 1998-124671	19980729
AU 9953245	A1	20000221	AU 1999-53245	19990728
EP 1100906	A1	20010523	EP 1999-938851	19990728

R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO

PRAI US 1998-124671 A 19980729
WO 1999-US17147 W 19990728
AB Inhibitors of the ***KDEL*** receptor that can be used to block the transfer of heat shock proteins to the endoplasmic reticulum and allow them to act as adjuvants are described. Certain proteins are functionally retained in the cellular endoplasmic reticulum via an interaction between a ***KDEL*** sequence and its receptor. According to the invention, blocking this interaction with a ***KDEL*** receptor inhibitor promotes the secretion of such proteins. In specific embodiments of the invention, ***KDEL*** receptor inhibitors may be used to promote the secretion of heat shock proteins, thereby rendering the secreted heat shock proteins more accessible to the immune system and improving the immune response to heat shock protein-assocd. antigens. The inhibitors are artificial peptides that oligomerize and present large no. of ***KDEL*** peptides to the receptors and sat. them. An example of one of these peptides uses the signal peptide of the BiP protein, an oligomerization domain of a cartilage oligomeric matrix protein, a linker peptide from a camel Ig and a ***KDEL*** peptide is described.
RE.CNT 2
RE
(1) Ciba Geigy Ag; WO 9818943 A 1998 CAPLUS
(2) Sloan-Kettering Institute For Cancer Research; WO 9706828 A 1997 CAPLUS

L41 ANSWER 9 OF 92 CAPLUS COPYRIGHT 2001 ACS
AN 2000:129323 CAPLUS
DN 132:275819
TI Retention of subunits of the oligosaccharyltransferase complex in the endoplasmic reticulum
AU Fu, Jie; Kreibich, Gert
CS Department of Cell Biology, New York University School of Medicine, New York, NY, 10016, USA
SO J. Biol. Chem. (2000), 275(6), 3984-3990
CODEN: JBCHA3; ISSN: 0021-9258
PB American Society for Biochemistry and Molecular Biology
DT Journal
LA English
AB Membrane proteins of the endoplasmic reticulum (ER) may be localized to this organelle by mechanisms that involve retention, retrieval, or a combination of both. For luminal ER proteins, which contain a ***KDEL*** domain, and for type I transmembrane proteins carrying a dilysine motif, specific retrieval mechanisms have been identified. However, most ER membrane proteins do not contain easily identifiable retrieval motifs. ER localization information has been found in cytoplasmic, transmembrane, or luminal domains. In this study, we have identified ER localization domains within the three type I transmembrane proteins, ribophorin I (RI), ribophorin II (RII), and OST48. Together with DAD1, these membrane proteins form an oligomeric complex that has oligosaccharyltransferase (OST) activity. We have previously shown that ER retention information is independently contained within the transmembrane and the cytoplasmic domain of RII, and in the case of RI, a truncated form consisting of the luminal domain was retained in the ER. To det. whether other domains of RI carry addnl. retention information, we have generated chimeras by exchanging individual domains of the Tac antigen with the corresponding ones of RI. We demonstrate here that only the luminal domain of RI contains ER retention information. We also show that the dilysine motif in OST48 functions as an ER localization motif because OST48 in which the two lysine residues are replaced by serine (OST48ss) is no longer retained in the ER and is found instead also at the plasma membrane. OST48ss is, however, retained in the ER when coexpressed with RI, RII, or chimeras, which by themselves do not exit from the ER, indicating that they may form partial oligomeric complexes by interacting with the luminal domain of OST48. In the case of the Tac chimera contg. only the luminal domain of RII, which by itself exits from the ER and is rapidly degraded, it is retained in the ER and becomes stabilized when coexpressed with OST48.
RE.CNT 56
RE
(1) Amar-Costesec, A; J Cell Biol 1984, V99, P2247 CAPLUS
(3) Bergeron, J; Trends Biochem Sci 1994, V19, P124 CAPLUS
(4) Biederer, T; EMBO J 1996, V15, P2069 CAPLUS
(5) Colley, K; J Biol Chem 1992, V267, P7784 CAPLUS
(6) Cosson, P; Curr Opin Cell Biol 1997, V9, P484 CAPLUS
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L41 ANSWER 10 OF 92 CAPLUS COPYRIGHT 2001 ACS
AN 2000:800591 CAPLUS
DN 134:70023
TI Production of hepatitis B surface antigen in transgenic plants for oral immunization
AU Richter, Liz J.; Thanavala, Yasmin; Arntzen, Charles J.; Mason, Hugh S.
CS Boyce Thompson Institutefor Plant Research, Inc, Ithaca, NY, 14853-1801, USA
SO Nat. Biotechnol. (2000), 18(11), 1167-1171
CODEN: NABIF9; ISSN: 1087-0156
PB Nature America Inc.
DT Journal
LA English
AB Here the authors present data showing oral immunogenicity of recombinant

hepatitis B surface antigen (HBsAg) in preclin. animal trials. Mice fed transgenic HBsAg potato tubers showed a primary immune response (increases in HBsAg-specific serum antibody) that could be greatly boosted by i.p. delivery of a single subimmunogenic dose of com. HBsAg vaccine, indicating that plants expressing HBsAg in edible tissues may be a new means for oral hepatitis B immunization. However, attainment of such a goal will require higher HBsAg expression than was obsd. for the potatoes used in this study. The authors conducted a systematic anal. of factors influencing the accumulation of HBsAg in transgenic potato, including 5' and 3' flanking elements and protein targeting within plant cells. The most striking improvements resulted from (1) alternative polyadenylation signals, and (2) ***fusion*** proteins contg. targeting signals designed to enhance integration or retention of HBsAg in the endoplasmic reticulum (ER) of plant cells.

RE.CNT 30

RE

(2) An, G; Plant Cell 1989, V1, P115 CAPLUS
(4) Becker, D; Plant Mol Biol 1992, V20, P1195 CAPLUS
(5) Bednarek, S; Plant Mol Biol 1992, V20, P133 CAPLUS
(6) Bruss, V; Intervirology 1996, V39, P23 CAPLUS
(7) Chan, M; Proc Natl Acad Sci USA 1998, V95, P6543 CAPLUS
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L41 ANSWER 11 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 4
AN 2001:22075 BIOSIS
DN PREV200100022075
TI Accumulation of maize gamma-zein and gamma-zein: ***KDEL*** to high levels in tobacco leaves and differential increase of BiP synthesis in transformants.
AU Bellucci, M.; Alpini, A.; Paolocci, E.; Cong, L.; Arcioni, S. (1)
CS (1) Istituto di Ricerche sul Miglioramento Genetico delle Piante Foraggere, CNR, Via Madonna Alta 130, 06128, Perugia: s.arcioni@irmgpf.pg.cnr.it Italy
SO Theoretical and Applied Genetics, (October, 2000) Vol. 101, No. 5-6, pp. 796-804. print.
ISSN: 0040-5752.
DT Article
LA English
SL English
AB Two gene constructs (pROK.TG1L and pROK.TG1LK) were utilized to achieve accumulation of maize gamma-zein to high levels in tobacco (Nicotiana tabacum L.) leaves. Both the chimaeric genes contained the gamma-zein-coding region preceded by the 5'untranslated leader from the coat protein mRNA of TMV, but one of them (pROK.TG1LK) was modified in its protein-coding region by the addition of the ER retention signal ***KDEL*** . The accumulation of gamma-zein and gamma-zein: ***KDEL*** in leaves was compared with ***heterologous*** protein accumulation in tobacco plants previously transformed with a gamma-zein cDNA harbouring a native 5'UTR. Replacement of gamma-zein 5'UTR with the TMV leader dramatically increased gamma-zein production. Furthermore, gamma-zein: ***KDEL*** -expressing plants, on average, accumulated twice as much foreign protein in their leaves as pROK.TG1L plants. The twofold increase in the level of gamma-zein: ***KDEL*** can probably be attributed to an improvement in the mechanism for ER retention of zeins in the transgenic cells. Transformants also showed increased production of BiP, though to a lesser extent in gamma-zein: ***KDEL*** -expressing plants compared with pROK.TG1L plants. It is therefore likely that gamma-zein: ***KDEL*** retention is made less dependent on the chaperone assistance of BiP by the presence of the ***KDEL*** signal on the gamma-zein mutant.

L41 ANSWER 12 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 5
AN 2000:240885 BIOSIS
DN PREV200000240885
TI Two distinct domains of the beta-subunit of glucosidase II interact with the catalytic alpha-subunit.
AU Arendt, Christopher W.; Ostergaard, Hanne L. (1)
CS (1) Department of Medical Microbiology and Immunology, University of Alberta, Edmonton, Alberta, T6G 2S2 Canada
SO Glycobiology, (May, 2000) Vol. 10, No. 5, pp. 487-492.
ISSN: 0959-6658.
DT Article
LA English
SL English
AB Recent purification and cDNA cloning of the endoplasmic reticulum processing enzyme glucosidase II have revealed that it is composed of two soluble proteins: a catalytic alpha-subunit and a beta-subunit of unknown function, both of which are highly conserved in mammals. Since the beta-subunit, which contains a C-terminal His-Asp-Glu-Leu (HDEL) motif, may function to link the catalytic subunit to the ***KDEL*** receptor as a retrieval mechanism, we sought to map the regions of the mouse beta-subunit protein responsible for mediating the association with the alpha-subunit. By screening a panel of recombinant beta-subunit glutathione S-transferase ***fusion*** proteins for the ability to precipitate glucosidase II activity, we have identified two non-overlapping interaction domains (ID1 and ID2) within the beta-subunit. ID1 encompasses 118 amino acids at the N-terminus of the mature polypeptide, spanning the cysteine-rich element in this region. ID2, located near the C-terminus, is contained within amino acids 273-400, a region occupied in part by a stretch of acidic residues. Variable usage of 7 alternatively spliced amino acids within ID2 was found not to influence the association of the two subunits. We theorize that the catalytic subunit of glucosidase II binds synergistically to ID1 and ID2, explaining the high associative stability of the enzyme complex.

L41 ANSWER 13 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 6
AN 2000:168095 BIOSIS
DN PREV200000168095
TI Development of ***chimeric*** molecules for recognition and targeting of antigen-specific B cells in pemphigus vulgaris.
AU Proby, C. M.; Ota, T.; Suzuki, H.; Koyasu, S.; Gamou, S.; Shimizu, N.; Wahl, J. K.; Wheelock, M. J.; Nishikawa, T.; Amagai, M. (1)
CS (1) Department of Dermatology, Keio University School of Medicine, 35 Shinanomachi, Shinjuku-ku, Tokyo, 160-8582 Japan
SO British Journal of Dermatology., (Feb., 2000) Vol. 142, No. 2, pp. 321-330.
ISSN: 0007-0963.
DT Article
LA English
SL English
AB Pemphigus vulgaris (PV) is an autoimmune blistering disease characterized by circulating pathogenic IgG antibodies against desmoglein 3 (Dsg3). The purpose of this study was to develop ***chimeric*** molecules for specific recognition and elimination of autoimmune B cells in PV. Mouse hybridoma cell lines producing anti-Dsg3 antibody (5H10, 12A2) were developed as an in vitro model system for targeting B cells. Dsg3-GFP, a baculoprotein containing the entire extracellular domain of Dsg3 fused with green fluorescence protein, recognized and targeted the hybridoma cells through their surface immunoglobulin receptors in an antigen-specific way. The epitopes of these monoclonal antibodies were mapped on the amino terminal EC1 and part of EC2, a region considered functionally important in cadherins. ***Chimeric*** toxin molecules containing the mapped region (Dsg3DELTAN1) and modified Pseudomonas exotoxin were produced in bacteria (Dsg3DELTAN1-PE40- ***KDEL*** , PE37-Dsg3DELTAN1- ***KDEL***) and tested in vitro on hybridoma cell lines. The ***chimeric*** toxins, but not Dsg3DELTAN1 alone, showed dose-dependent toxic activity with a reduction in hybridoma cell number to 40-60% of toxin-negative control cultures, compared with little or no effect on anti-Dsg3-negative hybridoma cells. Furthermore, these toxins showed toxic effects on anti-Dsg3 IgG-producing B cells from Dsg3DELTAN1-immunized mice, with a 60% reduction in cell number compared with Dsg3DELTAN1 alone. Thus, specific recognition and targeting of antigen-specific B cells in PV was demonstrated; this strategy may hold promise as a future therapeutic option for PV and other autoimmune diseases.

L41 ANSWER 14 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 7
AN 2001:127377 BIOSIS
DN PREV200100127377
TI Expression of maize gamma-zein and beta-zein genes in transgenic Nicotiana tabacum and Lotus corniculatus.
AU Bellucci, Michele; Alpini, Angelica; Arcioni, Sergio (1)
CS (1) Istituto di Ricerche sul Miglioramento Genetico delle Piante Foraggere (IRMGPF), CNR, Via Madonna Alta, 130, 06128, Perugia Italy
SO Plant Cell Tissue and Organ Culture, (2000) Vol. 62, No. 2, pp. 141-151. print.
ISSN: 0167-6857.
DT Article
LA English
SL English
AB Accumulation of zeins, the endosperm storage proteins of maize, in a ***heterologous*** plant expression system was attempted. Plants of Nicotiana tabacum and Lotus corniculatus were transformed by Agrobacterium with binary vectors harbouring genes that code for gamma-zein and beta-zein, two zeins rich in sulphur amino acids. Adding the ER retention signal ***KDEL*** to the C-terminal domain modified the zein polypeptides. Significant levels of gamma-zein: ***KDEL*** and beta-zein: ***KDEL*** were detected in primary transformants of tobacco. Moreover, the two zeins colocalized in leaf protein bodies of gamma-/beta-zein: ***KDEL*** plants derived from a cross between two primary transformants. Coexpression of gamma-zein: ***KDEL*** and beta-zein: ***KDEL*** could be a useful strategy to obtain genotypes of forage legumes which are able to accumulate sulphur amino acids to high levels. As a first step, L. corniculatus plants expressing gamma-zein: ***KDEL*** in the leaves were obtained.

L41 ANSWER 15 OF 92 CAPLUS COPYRIGHT 2001 ACS
AN 1999:753107 CAPLUS
DN 131:350254
TI Verotoxin B subunit for immunization
IN Green, Allan M.
PA USA
SO PCT Int. Appl., 47 pp.
CODEN: PIXXD2
DT Patent
LA English
FAN.CNT 1

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI WO 9959627	A2	19991125	WO 1999-US10679	19990514
WO 9959627	A3	20000120		

W: AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM

RW: GH, GM, KE, LS, MW, SD, SL, SZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG
AU 9939918 A1 19991206 AU 1999-39918 19990514
EP 1078007 A2 20010228 EP 1999-923063 19990514
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, FI
PRAI US 1998-85693 P 19980515
WO 1999-US10679 W 19990514
AB The author discloses methods for stimulating an immune response in a mammal by administering a toxin-antigen conjugate. In one example, a ***fusion*** construct of a MAGE-1 epitope and the B subunit of verotoxin was shown to undergo processing and MHC class I presentation by APC and to stimulate cytotoxic T-cells.

L41 ANSWER 16 OF 92 CAPLUS COPYRIGHT 2001 ACS
AN 2000:559644 CAPLUS
DN 133:131182
TI Insecticidal ***fusion*** protein, its coded gene and method for producing transgenosis strain using said gene
IN Zhu, Zhen; Deng, Chaoyang; Qu, Qiang
PA Genetics Inst., Chinese Academy of Sciences, Peop. Rep. China
SO Faming Zhuanli Shenqing Gongkai Shuomingshu, 55 pp.
CODEN: CNXXEV
DT Patent
LA Chinese
FAN.CNT 1

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI CN 1229087	A	19990922	CN 1999-103430	19990330

AB The disclosed insecticidal ***fusion*** protein contains signal peptide at its N-terminal, insecticidal protein, and endoplasmic reticulum-retention signal at its C-terminal. The signal peptide is selected from potato patatin signal peptide, pathogenesis-related protein PR signal peptide, and soybean Kunitz type trypsin inhibitor (SKTI) signal peptide; the insecticidal protein is selected from Bacillus thuringiensis (Bt) toxoprotein, cowpea trypsin inhibitor (CpTI) insect-resistant protein, paddy mercapto- protease inhibitor (OC), or bivalent insecticidal protein comprising their ***fusion*** proteins; and the signal peptide of the insecticidal protein and endoplasmic reticulum-retention signal such as ***KDEL*** and HDEL. The expression vector is a plant-transfecting vector, contains one or more insecticidal gene expression box and/or other gene expression box, and the exogenous gene of the expression box is controlled under plant promoter. The plant promoter is selected from CaMV 35S promoter, CLCuV replicase gene promoter, paddy actin promoter, T-DNA mas promoter, maize ubiquitin promoter, and their promoter complexes. The expression vector is used for prepn. of insect-resistant plants such as paddy, maize, wheat, tobacco, cotton, soybean, potato, cabbage, brassica oleracea, and pepper, etc. The transgenosis plant is prepd. by construction of expression vector encoding insecticidal ***fusion*** protein, transfecting plant cells with the vector, and culturing the plant cells.

L41 ANSWER 17 OF 92 EMBASE COPYRIGHT 2001 ELSEVIER SCI. B.V.DUPLICATE 8
AN 1999416801 EMBASE
TI Overexpression of cyclooxygenase-2 induces cell cycle arrest. Evidence for a prostaglandin-independent mechanism.
AU Trifan O.C.; Smith R.M.; Thompson B.D.; Hla T.
CS T. Hla, Center for Vascular Biology, Dept. of Physiology, Univ. of Connecticut Health Center, 263 Farmington Ave., Farmington, CT, United States. hla@sun.uchc.edu
SO Journal of Biological Chemistry, (26 Nov 1999) 274/48 (34141-34147). Refs: 29
ISSN: 0021-9258 CODEN: JBCHA3
CY United States
DT Journal; Article
FS 029 Clinical Biochemistry
037 Drug Literature Index
LA English
SL English
AB The immediate-early gene cyclooxygenase 2 (Cox-2) is induced in a variety of hyperplastic pathological conditions, including rheumatoid arthritis and colorectal cancer. Although a causal role for Cox-2 has been proposed, mechanisms by which Cox-2 function contributes to the pathogenesis of hyperplastic disease are not well defined. We constructed a green fluorescent protein-tagged Cox-2 (Cox-2-GFP) to examine its effects on a variety of cell types upon overexpression. Subcellular localization and enzymatic and pharmacological properties of Cox-2-GFP polypeptide were indistinguishable from those of the wild-type Cox-2 polypeptide. Overexpression of the Cox-2-GFP or the Cox-2 polypeptide by transient transfection suppressed the population of cells in the S phase of the cell cycle, with a concomitant increase in G0/G1 population. In contrast, transient overexpression of GFP had no effect on cell cycle distribution, whereas endoplasmic reticulum-retained GFP (GFP- ***KDEL***) overexpression was associated with only a minor decrease of cells in S phase. Interestingly, neither NS-398 (a Cox-2-specific inhibitor) nor indomethacin could reverse the effect of Cox-2-GFP overexpression on cell cycle progression. Furthermore, two mutants of Cox-2, S516Q and S516M, which lack the cyclooxygenase activity, exhibited the same effect as Cox-2-GFP. The cell cycle effect of Cox-2-GFP was observed in ECV-304, NIH 3T3, COS-7, bovine microvascular endothelial cells, and human embryonic kidney 293 cells. These findings suggest that Cox-2 inhibits cell cycle progression in a variety of cell types by a novel mechanism that does not require the synthesis of prostaglandins.

L41 ANSWER 18 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 9
AN 1999:442984 BIOSIS
DN PREV199900442984
TI Molecular characterization of a novel basement membrane-associated proteoglycan, leprecan.
AU Wassenhove-McCarthy, Deborah J.; McCarthy, Kevin J. (1)
CS (1) Dept. of Pathology, School of Medicine, Louisiana State University Medical Center, 1501 Kings Highway, Shreveport, LA, 71130 USA
SO Journal of Biological Chemistry, (Aug. 27, 1999) Vol. 274, No. 35, pp. 25004-25017.
ISSN: 0021-9258.
DT Article
LA English
SL English
AB A monoclonal antibody was used in early studies to identify a novel chondroitin sulfate proteoglycan, secreted by L-2 cells, the core protein of which was approximately 100 kDa. To characterize this proteoglycan core protein at the molecular level, an L-2 cell cDNA library was probed by expression screening and solution hybridization. Northern blot analysis assigned transcript size to approximately 3.1 kilobases and, after contig assembly, the coding region of the mRNA corresponded to 2.18 kilobases. Immunoassays were performed to confirm the identity of this sequence, using a polyclonal antibody raised against an expressed ***fusion*** protein encoded by sequence representing the carboxyl half of the molecule. The antibody recognized the core protein in Western blots after prior digestion of the intact proteoglycan with chondroitinase ABC. Immunostaining tissue sections with the same antibody localized the proteoglycan to basement membranes, and expression of the entire sequence in Chinesehamster ovary K-1 cells showed that the protein encoded by the sequence secreted as a chondroitin sulfate proteoglycan. The core protein not only has motifs permitting glycosylation as a proteoglycan, but also possesses the endoplasmic reticulum retrieval signal, ***KDEL*** , which suggests that, in addition to its role as a basement membrane component, it may also participate in the secretory pathway of cells.

L41 ANSWER 19 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 10
AN 1999:134298 BIOSIS
DN PREV199900134298
TI Structural basis for the differential toxicity of cholera toxin and Escherichia coli heat-labile enterotoxin. Construction of ***hybrid*** toxins identifies the A2-domain as the determinant of differential toxicity.
AU Rodighiero, Chiara; Aman, Abu T.; Kenny, Martin J.; Moss, Joel; Lencer, Wayne I.; Hirst, Timothy R. (1)
CS (1) Dep. Pathol. Microbiol., Univ. Bristol, Sch. Med. Sci., University Walk, Bristol BS8 1TD UK
SO Journal of Biological Chemistry, (Feb. 12, 1999) Vol. 274, No. 7, pp. 3962-3969.
ISSN: 0021-9258.
DT Article
LA English
AB Cholera toxin (Ctx) and E. coli heat-labile enterotoxin (Etx) are structurally and functionally similar AB5 toxins with over 80% sequence identity. When their action in polarized human epithelial (T84) cells was monitored by measuring toxin-induced Cl- ion secretion, Ctx was found to be the more potent of the two toxins. Here, we examine the structural basis for this difference in toxicity by engineering a set of mutant and ***hybrid*** toxins and testing their activity in T84 cells. This revealed that the differential toxicity of Ctx and Etx was (i) not due to differences in the A-subunit's C-terminal ***KDEL*** targeting motif (which is RDEL in Etx), as a Y.DEL to RDEL substitution had no effect on cholera toxin activity; (ii) not attributable to the enzymatically active A1-fragment, as ***hybrid*** toxins in which the A1-fragment in Ctx was substituted for that of Etx (and vice versa) did not alter relative toxicity; and (iii) not due to the B-subunit, as the replacement of the B-subunit in Ctx for that of Etx caused no alteration in toxicity, thus excluding the possibility that the broader receptor specificity of EtxB is responsible for reduced activity. Remarkably, the difference in toxicity could be mapped to a 10-amino acid segment of the A2-fragment that penetrates the central pore of the B-subunit pentamer. A comparison of the in vitro stability of two ***hybrid*** toxins, differing only in this 10-amino acid segment, revealed that the Ctx A2-segment conferred a greater stability to the interaction between the A- and B-subunits than the corresponding segment from Etx A2. This suggests that the reason for the relative potency of Ctx compared with Etx stems from the increased ability of the A2-fragment of Ctx to maintain holotoxin stability during uptake and transport into intestinal epithelia.

L41 ANSWER 20 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 11
AN 1999:210816 BIOSIS
DN PREV199900210816
TI The transmembrane domain of hepatitis C virus glycoprotein E1 is a signal for static retention in the endoplasmic reticulum.
AU Cocquerel, Laurence; Duvet, Sandrine; Meunier, Jean-Christophe; Pillez, Andre; Cacan, Rene; Wychowski, Czeslaw; Dubuisson, Jean (1)
CS (1) Equipe Hepatite C, CNRS-UMR 319, Institut de Biologie de Lille and Institut Pasteur de Lille, 1 rue Calmette, 59021, Lille Cedex France
SO Journal of Virology, (April, 1999) Vol. 73, No. 4, pp. 2641-2649.
ISSN: 0022-538X.
DT Article

LA English
SL English
AB Hepatitis C virus (HCV) glycoproteins E1 and E2 assemble to form a noncovalent heterodimer which, in the cell, accumulates in the endoplasmic reticulum (ER). Contrary to what is observed for proteins with a ***KDEL*** or a KKXX ER-targeting signal, the ER localization of the HCV glycoprotein complex is due to a static retention in this compartment rather than to its retrieval from the cis-Golgi region. A static retention in the ER is also observed when E2 is expressed in the absence of E1 or for a ***chimeric*** protein containing the ectodomain of CD4 in ***fusion*** with the transmembrane domain (TMD) of E2. Although they do not exclude the presence of an intracellular localization signal in E1, these data do suggest that the TMD of E2 is an ER retention signal for HCV glycoprotein complex. In this study ***chimeric*** proteins containing the ectodomain of CD4 or CD8 fused to the C-terminal hydrophobic sequence of E1 were shown to be localized in the ER, indicating that the TMD of E1 is also a signal for ER localization. In addition, these ***chimeric*** proteins were not processed by Golgi enzymes, indicating that the TMD of E1 is responsible for true retention in the ER, without recycling through the Golgi apparatus. Together, these data suggest that at least two signals (TMDs of E1 and E2) are involved in ER retention of the HCV glycoprotein complex.

L41 ANSWER 21 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 12
AN 2000:50138 BIOSIS
DN PREV200000050138
TI Rab6 coordinates a novel Golgi to ER retrograde transport pathway in live cells.
AU White, Jamie (1); Johannes, Ludger; Mallard, Frederic; Girod, Andreas; Grill, Stephan; Reinsch, Sigrid; Keller, Patrick; Tzschaschel, Barbara; Echard, Arnaud; Goud, Bruno; Stelzer, Ernst H. K.
CS (1) Light Microscopy Group and Cell Biophysics and Cell Biology Programme, European Molecular Biology Laboratory, Meyerhofstrasse 1, D-69117, Heidelberg Germany
SO Journal of Cell Biology, (Nov. 15, 1999) Vol. 147, No. 4, pp. 743-759. ISSN: 0021-9525.
DT Article
LA English
SL English
AB We visualized a fluorescent-protein (FP) ***fusion*** to Rab6, a Golgi-associated GTPase, in conjunction with fluorescent secretory pathway markers. FP-Rab6 defined highly dynamic transport carriers (TCs) translocating from the Golgi to the cell periphery. FP-Rab6 TCs specifically accumulated a retrograde cargo, the wild-type Shiga toxin B-fragment (STB), during STB transport from the Golgi to the endoplasmic reticulum (ER). FP-Rab6 TCs associated intimately with the ER, and STB entered the ER via specialized peripheral regions that accumulated FP-Rab6. Microinjection of antibodies that block coatomer protein I (COPI) function inhibited trafficking of a ***KDEL*** -receptor FP-***fusion*** , but not FP-Rab6. Additionally, markers of COPI-dependent recycling were excluded from FP-Rab6/STB TCs. Overexpression of Rab6:GDP (T27N mutant) using T7 vaccinia inhibited toxicity of Shiga holotoxin, but did not alter STB transport to the Golgi or Golgi morphology. Taken together, our results indicate Rab6 regulates a novel Golgi to ER transport pathway.

L41 ANSWER 22 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 13
AN 2000:50319 BIOSIS
DN PREV200000050319
TI Calreticulin is transported to the surface of NG108-15 cells where it forms surface patches and is partially degraded in an acidic compartment.
AU Xiao, Guangqing; Chung, Tzu-Feng; Fine, Richard E.; Johnson, Robin J. (1)
CS (1) Department of Biochemistry, Boston University School of Medicine, Boston, MA USA
SO Journal of Neuroscience Research, (Dec. 1, 1999) Vol. 58, No. 5, pp. 652-662. ISSN: 0360-4012.
DT Article
LA English
SL English
AB Although calreticulin (Crt) is primarily localized to the endoplasmic reticulum (ER), our results using biotinylation and immunocytochemical methods indicate that a small but significant amount of Crt is present and forms large patches on the surface of NG108-15 cells (a mouse neuroblastoma-rat glioma ***hybrid*** cell line). 35S-labelled Crt molecules begin to reach the cell surface after only 10 min of labelling and disappear slowly from the cell surface. After 4 hr of labelling, approximately half of the newly synthesized Crt molecules are on the cell surface. We believe that some Crt molecules may escape from the ***KDEL*** receptor-mediated salvage pathway as they are synthesized and proceed through the secretory pathway to the cell surface. Immunoprecipitation from the culture medium shows that Crt is not released from the cell surface to the medium, suggesting tight binding to surface molecules. NH4Cl can block the degradation of Crt; therefore, Crt is presumably degraded in the lysosome pathway. However, blockage of the disappearance of surface Crt is less efficient than that of internal Crt. This suggests that the disappearance of Crt from the cell surface may not be due solely to its degradation, but may reflect transport into another cell compartment such as the ER.

L41 ANSWER 23 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 14
AN 1999:342870 BIOSIS
DN PREV199900342870
TI Morphological and functional association of Sec22b/ERS-24 with the pre-Golgi intermediate compartment.
AU Zhang, Tao; Wong, Siew Heng; Tang, Bor Luen; Xu, Yue; Hong, Wanjin (1)
CS (1) Membrane Biology Laboratory, Institute of Molecular and Cell Biology, Singapore, 117609 Singapore
SO Molecular Biology of the Cell, (Feb., 1999) Vol. 10, No. 2, pp. 435-453. ISSN: 1059-1524.
DT Article
LA English
SL English
AB Yeast Sec22p participates in both anterograde and retrograde vesicular transport between the endoplasmic reticulum (ER) and the Golgi apparatus by functioning as a v-SNARE (soluble N-ethylmaleimide-sensitive factor (NSF) attachment protein receptor) of transport vesicles. Three mammalian proteins homologous to Sec22p have been identified and are referred to as Sec22a, Sec22b/ERS-24, and Sec22c, respectively. The existence of three homologous proteins in mammalian cells calls for detailed cell biological and functional examinations of each individual protein. The epitope-tagged forms of all three proteins have been shown to be primarily associated with the ER, although functional examination has not been carefully performed for any one of them. In this study, using antibodies specific for Sec22b/ERS-24, it is revealed that endogenous Sec22b/ERS-24 is associated with vesicular structures in both the perinuclear Golgi and peripheral regions. Colabeling experiments for Sec22b/ERS-24 withGolgi mannosidase II, the ***KDEL*** receptor, and the envelope glycoprotein G (VSVG) of vesicular stomatitis virus (VSV) en route from the ER to the Golgi under normal, brefeldin A, or nocodazole-treated cells suggest that Sec22b/ERS-24 is enriched in the pre-Golgi intermediate compartment (IC). In a well-established semi-intact cell system that reconstitutes transport from the ER to the Golgi, transport of VSVG is inhibited by antibodies against Sec22b/ERS-24. EGTA is known to inhibit ER-Golgi transport at a stage after vesicle/transport intermediate docking but before the actual ***fusion*** event. Antibodies against Sec22b/ERS-24 inhibit ER-Golgi transport only when they are added before the EGTA-sensitive stage. Transport of VSVG accumulated in pre-Golgi IC by incubation at 15degreeC is also inhibited by Sec22b/ERS-24 antibodies. Morphologically, VSVG is transported from the ER to the Golgi apparatus via vesicular intermediates that scatter in the peripheral as well as the Golgi regions. In thepresence of antibodies against Sec22b/ERS-24, VSVG is seen to accumulate in these intermediates, suggesting that Sec22b/ERS-24 functions at the level of the IC in ER-Golgi transport.

L41 ANSWER 24 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 15
AN 1999:96922 BIOSIS
DN PREV199900096922
TI Alternative mechanisms of interaction between homotypic and heterotypic parainfluenza virus HN and F proteins.
AU Tong, Suxiang; Compans, Richard W. (1)
CS (1) Dep. Microbiol. Immunol., Emory Univ., Atlanta, GA 30322 USA
SO Journal of General Virology, (Jan., 1999) Vol. 80, No. 1, pp. 107-115. ISSN: 0022-1317.
DT Article
LA English
AB Cell ***fusion*** by human parainfluenza virus (HPIV) type 2 or type 3 requires the coexpression of both the ***fusion*** (F) and haemagglutinin-neuraminidase (HN) glycoproteins from the same virus type, indicating that promotion of ***fusion*** requires a type-specific interaction between F and HN. In this report we have further investigated the interaction of the ectodomains of the F and HN glycoproteins from HPIV2 and HPIV3. We constructed mutants of the HPIV2 F and HPIV3 F proteins (F'- ***KDEL***) lacking a transmembrane anchor and a cytoplasmic tail, and containing a C-terminal signal for retention in the endoplasmic reticulum (ER). The P12 and P13 F'- ***KDEL*** proteins were both found to be retained intracellularly, and neither could induce cell ***fusion*** when co-expressed with homotypic HN proteins. Qualitative and quantitative cell- ***fusion*** assays also showed that both the P12 F'- ***KDEL*** and P13 F'- ***KDEL*** proteins have inhibitory effects on P12 F- and HN-induced cell ***fusion*** . However, the F- ***KDEL*** mutants were found to inhibit cell ***fusion*** by two distinct mechanisms. An interaction between P12 F'- ***KDEL*** and P12 HN results in intracellular retention of HN, and a block in its transport to the cell surface. In contrast, P13 F'- ***KDEL*** was found to suppress the steady-state intracellular expression levels of HPIV2 HN. These results support the conclusion that ***fusion*** involves an interaction between the HN and F proteins, and suggest that an association between F and HN may occur in the ER.

L41 ANSWER 25 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 16
AN 2000:30339 BIOSIS
DN PREV200000030339
TI Accumulation of antibody ***fusion*** proteins in the cytoplasm and ER of plant cells.
AU Spiegel, Holger; Schillberg, Stefan (1); Sack, Markus; Holzem, Achim; Naehring, Joerg; Monecke, Michael; Liao, Yu-Cai; Fischer, Rainer
CS (1) Institut fuer Biologie I (Botanik/Molekulargenetik), RWTH Aachen, Worringerweg 1, D-52074, Aachen Germany
SO Plant Science (Shannon), (Nov. 12, 1999) Vol. 149, No. 1, pp. 63-71. ISSN: 0168-9452.
DT Article
LA English
SL English
AB To test whether the accumulation of cytoplasmically targeted recombinant antibodies could be improved by ***fusion*** to a cytoplasmic protein,

we generated a series of single chain antibody- ***fusion*** proteins and assayed the levels of functional protein. Glutathione S-transferase (GST) from Schistosoma japonicum, coat protein (CP) from TMV, thioredoxin from tobacco (TRXt) or thioredoxin from Escherichia coli (TRXe) was fused to the N-terminus of scFv24, a TMV specific single chain antibody. Accumulation of functional ***fusion*** proteins in the endoplasmic reticulum (ER) and plant cell cytoplasm was analysed by transient expression in tobacco leaves. ELISA analysis demonstrated that the ***fusion*** partners did not prevent the binding of scFv24 to TMV virions. However, accumulation of functional scFv24 was dependent on the ***fusion*** partner coupled to it. CP-scFv and GST-scFv ***fusion*** protein accumulation amounted to 1 mug and 3 mug/g of leaf material, respectively, whereas the thioredoxin ***fusion*** proteins were produced at low levels. Western blot and surface plasmon resonance analysis confirmed the integrity of the ER retained CP and GST ***fusion*** proteins. In the cytoplasm, only the CP ***fusion*** protein was detectable (1-5 ng/gram of leaf material) and levels of scFv24 alone or fused to the other three ***fusion*** partners were below the ELISA detection limit. Addition of a ***KDEL*** sequence to the C-terminus of the cytoplasmic CP ***fusion*** resulted in a 3-fold increase in protein accumulation indicating that an N-terminal CP and the C-terminal ***KDEL*** sequence are suitable elements to stabilize scFv antibodies in the cytoplasm.

L41 ANSWER 26 OF 92 CAPLUS COPYRIGHT 2001 ACS
AN 1998:568850 CAPLUS
DN 129:185085
TI Modified prodomain C-terminus of human carboxypeptidase B that enhances recombinant expression of the mature enzyme
IN Edge, Michael Derek
PA Zeneca Limited, UK
SO PCT Int. Appl., 88 pp.
CODEN: PIXXD2
DT Patent
LA English
FAN.CNT 1

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI WO 9835988	A1	19980820	WO 1998-GB415	19980210
W: AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GE, GH, GM, GW, HU, ID, IL, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, US, UZ, VN, YU, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, SD, SZ, UG, ZW, AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG				
AU 9860006	A1	19980908	AU 1998-60006	19980210
PRAI GB 1997-3104		19970214		
GB 1997-22003		19971018		
GB 1997-22727		19971029		
WO 1998-GB415		19980210		

AB The field of the invention is recombinant prodn. of carboxypeptidase B. This invention provides a modified prodomain of carboxypeptidase B which enhances recombinant expression thereof when co-expressed from a sep. gene. Preferred modified prodomains (residues 1-95 of the proenzyme) have added amino acids at their C-terminus, in particular any one of the following sequences: L, ***KDEL*** , KKAA or SDYQRL. The carboxypeptidase is preferably human pancreatic carboxypeptidase B. The invention also relates to corresponding polynucleotide sequences, vectors, host cells and methods of recombinant carboxypeptidase B prodn. Expression of mature human pancreatic carboxypeptidase B from COS cells is enhanced by co-secretion of the modified prodomain. An esp. preferred carboxypeptidase B ***fusion*** construct comprises a gene encoding a humanized Fd heavy chain fragment of antibody 806.077 linked to [A248S,G251T,D253K]-human carboxypeptidase B and its co-expression with a gene encoding a humanized light chain of 806.077 and a gene encoding the pro-Leu modified prodomain of human carboxypeptidase B to give the F(ab')2 protein with a mol. of [A248S,G251T,D253K]carboxypeptidase B at the C-terminus of each of the heavy chain fragments. The const. and hinge regions of the humanized Fd heavy chain fragment are derived from the human IgG3 antibody type.

L41 ANSWER 27 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 17
AN 1998:386332 BIOSIS
DN PREV199800386332
TI Interaction between a Ca2+-binding protein calreticulin and perforin, a component of the cytotoxic T-cell granules.
AU Andrin, Christi; Pinkoski, Michael J.; Burns, Kimberly; Atkinson, Eric A.; Krahenbuhl, Olivier; Hudig, Dorothy; Fraser, Stephanie A.; Winkler, Urlike; Tschopp, Juerg; Opas, Michal; Bleackley, R. Chris; Michalak, Marek (1)
CS (1) Mol. Biol. Membranes Res. Group, Univ. Alberta, AB T6G 2H7 Canada
SO Biochemistry, (July 21, 1998) Vol. 37, No. 29, pp. 10386-10394.
ISSN: 0006-2960.
DT Article
LA English
AB Calreticulin is a component of cytotoxic T-lymphocyte and NK lymphocyte granules. We report here that granule-associated calreticulin terminates with the ***KDEL*** endoplasmic reticulum retrieval amino acid sequence and somehow escapes the ***KDEL*** retrieval system. In perforin knock-out mice calreticulin is still targeted into the granules. Thus, calreticulin will traffic without perforin to cytotoxic granules. In the granules, calreticulin and perforin are associated as documented by (i) copurification of calreticulin with perforin but not with granzymes and (ii) immunoprecipitation of a calreticulin-perforin complex using specific antibodies. By using calreticulin affinity chromatography and protein ligand blotting we show that perforin binds to calreticulin in the absence of Ca2+ and the two proteins dissociate upon exposure to 0.1 mM or higher Ca2+ concentration. Perforin interacts strongly with the P-domain of calreticulin (the domain which has high Ca2+-binding affinity and chaperone function) as revealed by direct protein-protein interaction, ligand blotting, and the yeast two- ***hybrid*** techniques. Our results suggest that calreticulin may act as Ca2+-regulated chaperone for perforin. This action will serve to protect the CTL during biogenesis of granules and may also serve to regulate perforin lytic action after release.

L41 ANSWER 28 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 18
AN 1998:394620 BIOSIS
DN PREV199800394620
TI Modulation of apoptotic response of a radiation-resistant human carcinoma by Pseudomonas exotoxin- ***chimeric*** protein.
AU Seetharam, Saraswathy; Nodzenski, Edwardine; Beckett, Michael A.; Heimann, Ruth; Cha, Amy; Margulies, Inger; Pastan, Ira; Kufe, Donald W.; Weichselbaum, Ralph R. (1)
CS (1) Dep. Radiation Cell. Oncol., Univ. Chicago Hosp., Chicago, IL 60637 USA
SO Cancer Research, (Aug. 1, 1998) Vol. 58, No. 15, pp. 3215-3220.
ISSN: 0008-5472.
DT Article
LA English
AB Strategies to sensitize human tumors that an resistant to apoptosis have been clinically unsuccessful. We demonstrate that a structurally modified ***chimeric*** Pseudomonas exotoxin, PEDELTA53L/TGF-alpha/ ***KDEL*** , with binding specificity for the epidermal growth factor receptor, markedly enhances sensitivity of human xenografts to radiation killing. Exposure to PEDELTA53L/TGF-alpha/ ***KDEL*** decreases the apoptotic threshold through protein synthesis inhibition and simultaneous production of ceramide in tumor cells that lack functional p53 protein. In contrast, no increase in local or systemic toxicity was observed with the ***chimeric*** toxin and radiation. We conclude that biochemical targeting of the ***chimeric*** toxin and physical targeting of ionizing radiation may increase the therapeutic ratio in the treatment of human cancers with alterations of p53 expression. This strategy offers a high therapeutic potential for Pseudomonas exotoxin A ***chimeric*** proteins and irradiation.

L41 ANSWER 29 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 19
AN 1998:480491 BIOSIS
DN PREV199800480491
TI Major histocompatibility complex class I presentation of exogenous soluble tumor antigen fused to the B-fragment of Shiga toxin.
AU Lee, Ren-Shiang; Tartour, Eric (1); Van Der Bruggen, Pierre; Vantomme, Valerie; Joyeux, Isabelle; Goud, Bruno; Fridman, Wolf Herman; Johannes, Ludger
CS (1) Lab. Immunol. Clin., INSERM U255, Inst. Curie, 26 rue Ulm, F-75248 Paris Cedex 05 France
SO European Journal of Immunology, (Sept., 1998) Vol. 28, No. 9, pp. 2726-2737.
ISSN: 0014-2980.
DT Article
LA English
AB Targeting exogenous antigen into the MHC class I-restricted presentation pathway is a prerequisite for the induction of cytotoxic T lymphocytes (CTL) which. have been shown to represent an important component of the protective and therapeutic immune response to viral infections and tumors. In this study, we produced recombinant proteins composed of the receptor-binding non-toxic B-fragment of bacterial Shiga toxin derived from Shigella dysenteriae associated with an epitope from a model tumor antigen, Mage 1. We show that Shiga B-Mage 1 ***fusion*** proteins carrying an active or inactive endoplasmic reticulum retrieval signal (the C-terminal peptides ***KDEL*** or KDELGL, respectively) could be presented by peripheral blood mononuclear cells in an MHC class I-restricted manner to Mage 1-specific CTL. After pulsing B lymphoblastoid cells or dendritic cells with-Shiga B-Mage-1 ***fusion*** -protein, activation of the MHC class I-restricted Mage 1-specific CTL was also demonstrated. In further analysis, we showed that treatment with brefeldin A or paraformaldehyde fixation of Epstein-Barr virus-transformed B cells prevented the presentation of the Mage 1 T cell epitope, which excluded extracellular processing of the antigen. Immunofluorescence analysis also revealed that the Shiga B-Mage 1 ***fusion*** protein was largely excluded from Lamp-2-positive lysosomal structures. Therefore, the ability of Shiga toxin B-fragment to target dendritic cells and B cells and to direct antigen into the exogenous class I-restricted pathway makes it an attractive non-living and non-toxic vaccine vector.

L41 ANSWER 30 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 20
AN 1998:443391 BIOSIS
DN PREV199800443391
TI Differences in cytotoxicity of native and engineered RIPs can be used to assess their ability to reach the cytoplasm.
AU Svinth, Maria; Steighardt, Jorg; Hernandez, Raquel; Suh, Jung-Keun; Kelly, Curtis; Day, Philip; Lord, Michael; Girbes, Tomas; Robertus, Jon D. (1)
CS (1) Inst. Cellular Mol. Biol., Dep. Chem. Biochem., Univ. Tex., Austin, TX

78712 USA
SO Biochemical and Biophysical Research Communications, (Aug. 28, 1998) Vol. 249, No. 3, pp. 637-642.
ISSN: 0006-291X.
DT Article
LA English
AB Ricin is a heterodimeric cytotoxin composed of RTB, a galactose binding lectin, and RTA, an enzymatic N-glycosidase. The toxin is endocytosed, and after intracellular routing, RTA is translocated to the cytoplasm where it inactivates ribosomes resulting in a loss of host cell protein synthesis and cell death. We show for the first time that the cytotoxicity against cultured T cells by several RTA mutants is directly proportional to the enzyme activity of RTA, suggesting this is a reliable system to measure translocation effects. Large discrepancies between cytotoxicity and enzyme action for a given pair of toxins are therefore attributable to differences in cell binding, uptake, or membrane translocation. Fluid phase uptake and cytotoxicity of isolated RTA are essentially identical to that of the single chain toxin PAP. This important finding suggests that RTA, and the A chain of class 2 RIPs in general, has not evolved special translocation signals to complement the increased target cell binding facilitated by RTB. Experiments with the lectin RCA and with ebulin suggest those toxins have diminished cytotoxicity probably mediated by comparative deficiencies in B chain binding. Addition of a ***KDEL*** sequence to RTA increases fluid phase uptake, consistent with the notion that transport to the ER is important for cytotoxicity. ***Fusion*** of MBP or GST to the amino terminus of RTA has little effect on enzyme action or cytotoxicity. This result is not altered by protease inhibitors, suggesting the ***fusion*** proteins are probably not cleaved prior to translocation of the toxic A chain and implying that the toxins can carry large passenger proteins into the cytoplasm, an observation with interesting potential for analytical and therapeutic chemistry.

L41 ANSWER 31 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 21
AN 1998:166687 BIOSIS
DN PREV199800166687
TI Design, characterization and anti-tumour cytotoxicity of a panel of recombinant, mammalian ribonuclease-based immunotoxins.
AU Deonarain, M. P. (1); Epenetos, A. A.
CS (1) Dep. Biochem., Imperial Coll. Sci. Technol. Med., London SW7 2AY UK
SO British Journal of Cancer, (Feb., 1998) Vol. 77, No. 4, pp. 537-546.
ISSN: 0007-0920.
DT Article
LA English
AB Bovine seminal ribonuclease (BSRNase) is an unusual member of the ribonuclease superfamily, because of its remarkable antitumour and immunosuppressive properties. We describe here the construction, expression, purification and characterization of a panel of six immunotoxins based upon this enzyme and show that we can increase its anti-tumour activity by over 2 X 104-fold. This is achieved by improving tumour cell targeting using a single-chain Fv (scFv) directed against the oncofetal antigen placental alkaline phosphatase. As well as the simple scFv-BSRNase ***fusion*** protein, we have constructed five other derivatives with additional peptides designed to improve folding and intracellular trafficking and delivery. We find that the molecule most cytotoxic to antigen (PLAP)-positive cells in vitro is one that contains a C-terminal ' ***KDEL*** ' endoplasmic reticulum retention signal and a peptide sequence derived from diphtheria toxin. All these molecules are produced in Escherichia coli (E. coli) as insoluble inclusion bodies and require extensive in vitro processing to recover antigen binding and ribonuclease activity. Despite incomplete ribonuclease activity and quaternary assembly, these molecules are promising reagents for specific chemotherapy of cancer and are potentially less harmful and immunogenic than current immunotoxins.

L41 ANSWER 32 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 22
AN 1998:402170 BIOSIS
DN PREV199800402170
TI Stacks on tracks: The plant Golgi apparatus traffics on an actin/ER network.
AU Boevink, Petra; Oparka, Karl; Cruz, Simon Santa; Martin, Barry; Betteridge, Alan; Hawes, Chris (1)
CS (1) Res. Sch. Biol. Mol. Sci., Oxford Brookes Univ., Gipsy Lane, Oxford OX3 0BP UK
SO Plant Journal, (Aug., 1998) Vol. 15, No. 3, pp. 441-447.
ISSN: 0960-7412.
DT Article
LA English
AB We have visualized the relationship between the endoplasmic reticulum (ER) and Golgi in leaf cells of Nicotiana clevelandii by expression of two Golgi proteins fused to green fluorescent protein (GFP). A ***fusion*** of the transmembrane domain (signal anchor sequence) of a rat sialyl transferase to GFP was targeted to the Golgi stacks. A second construct that expressed the Arabidopsis H/ ***KDEL*** receptor homologue aERD2, fused to GFP, was targeted to both the Golgi apparatus and ER, allowing the relationship between these two organelles to be studied in living cells for the first time. The Golgi stacks were shown to move rapidly and extensively along the polygonal cortical ER network of leaf epidermal cells, without departing from the ER tubules. Co-localization of F-actin in the GFP-expressing cells revealed an underlying actin cytoskeleton that matched precisely the architecture of the ER network, while treatment of cells with the inhibitors cytochalasin D and N-ethylmaleimide revealed the dependency of Golgi movement on actin cables. These observations suggest that the leaf Golgi complex functions as a motile system of actin-directed stacks whose function is to pick up products from a relatively stationary ER system. Also, we demonstrate for the first time in vivo brefeldin A-induced retrograde transport of Golgi membrane protein to the ER.

L41 ANSWER 33 OF 92 CAPLUS COPYRIGHT 2001 ACS
AN 1999:36136 CAPLUS
DN 130:219334
TI Differential activity of cholera toxin and E. coli enterotoxin: construction and purification of mutant and ***hybrid*** derivatives
AU Rodighiero, C.; Aman, A. T.; Lencer, W. I.; Hirst, T. R.
CS Department of Pathology and Microbiology, School of Medical Sciences, University of Bristol, Bristol, BS8 1TD, UK
SO Biochem. Soc. Trans. (1998), 26(4), S364
CODEN: BCSTB5; ISSN: 0300-5127
PB Portland Press Ltd.
DT Journal
LA English
AB To det. whether the differential toxicity of cholera toxin (Ctx) and Escherichia enterotoxin (Etx) lies within the A- or B- subunits of the mols., chimeras have been engineered which comprise portions of the A-subunit of Ctx complexed with the B-subunit of Etx and vice versa. A mutant cholera toxin in which the C-terminal ER retention signal (***KDEL***) was substituted for RDEL found in Etx, was also prepd. Here the authors describe the genetic construction of mutant and ***hybrid*** toxins and a method for their purifn.
RE.CNT 6
RE
(1) Amin, T; Prot Expr Purif 1994, V5, P198 CAPLUS
(2) Hirst, T; Handbook of Natural Toxins 1995, V8, P123 CAPLUS
(3) Kaper, J; Nature 1984, V308, P655 CAPLUS
(4) Lencer, W; J Cell Biol 1995, V131, P951 CAPLUS
(5) Mekalanos, J; Meth Enzym 1988, V165, P169 CAPLUS
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L41 ANSWER 34 OF 92 CAPLUS COPYRIGHT 2001 ACS
AN 1998:588578 CAPLUS
DN 129:300572
TI Localization of endoplasmic reticulum in living cells using green fluorescent protein chimeras
AU Van Goethem, Iris D. A.; Adams, Phil; Chad, John E.; Mather, Andrea M.; Griffiths, Barbara; Lee, Anthony G.; East, J. Malcolm
CS School of Biological Sciences, Department of Biochemistry and Molecular Biology, University of Southampton, Southampton, SO167PX, UK
SO Biochem. Soc. Trans. (1998), 26(3), S298
CODEN: BCSTB5; ISSN: 0300-5127
PB Portland Press Ltd.
DT Journal
LA English
AB In order to examine the location of sarcoplasmic/endoplasmic calcium pumps (SERCAs) in COS 7 cells a chimera of SERCA1a and green fluorescent protein (GFP) of Aequorea victoria was produced. In order to det. the location of endoplasmic reticulum (ER) a construct contg. the ER targeting sequence from .alpha.1-antitrypsin attached to GFP terminating with the ER retrieval sequence (***KDEL***) (designated GAP-K) was produced. In order to be certain that the SERCA1a-GFP ***fusion*** protein is correctly targeted the calcium transport properties of the chimera were characterized. SERCA1a-GFP and GAP-K occupied similar internal membrane compartments, presumably ER. A comparison of SERCA1a and SERCA1a-GFP localization indicated that the addn. of GFP to the C-terminus of SERCA1a had not altered its cellular location. The finding that SERCA1a-GFP is able to pump calcium make it unlikely that the ER location of the ***fusion*** protein is the result of mis-folding.

L41 ANSWER 35 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 23
AN 1998:165983 BIOSIS
DN PREV199800165983
TI Cloning and expression of two genes encoding auxin-binding proteins from tobacco.
AU Watanabe, Shinichiro; Shimomura, Shoji (1)
CS (1) Natl. Inst. Agrobiological Resources, Kannondai 2-1-2, Tsukuba, Ibaraki, 305 Japan
SO Plant Molecular Biology, (Jan. 1, 1998) Vol. 36, No. 1, pp. 63-74.
ISSN: 0167-4412.
DT Article
LA English
AB Two genes encoding the auxin-binding protein (ABP1) of tobacco (Nicotiana tabacum L.), both of which possess the characteristics of a luminal protein of the endoplasmic reticulum (ER), were isolated and sequenced. These genes were composed of at least five exons and four introns. The two coding exons showed 95% sequence homology and coded for two precursor proteins of 187 amino acid residues with molecular masses of 21 256 and 21 453 Da. The deduced amino acid sequences were 93% identical and both possessed an amino-terminal signal peptide, a hydrophilic mature protein region with two potential N-glycosylation sites and a carboxyl-terminal sorting signal, ***KDEL*** , for the ER. Restriction mapping of the cDNAs encoding tobacco ABP1, previously purified by amplification of tobacco cDNA libraries by polymerase chain reaction (PCR) using specific primers common to both genes, indicated that both genes were expressed, although one was expressed at a higher level than the other. Genomic Southern blot hybridization showed no other homologous genes except for these two in the tobacco genome. The apparent molecular mass of the mature

form of tobacco ABP1 was revealed to be 25 kDa by SDS polyacrylamide gel electrophoresis using affinity-purified anti (tobacco ABP1) antibodies raised against a ***fusion*** protein with maltose-binding protein. Expression of the recombinant ABP1 gene in transgenic tobacco resulted in accumulation of the 25 kDa protein. A single point mutation of an amino acid residue at either of the two potential N-glycosylation sites resulted in a decrease in the apparent molecular mass and produced a 22 kDa protein. Mutations at both sites resulted in the formation of a 19.3 kDa protein, suggesting that tobacco ABP1 is glycosylated at two asparagine residues.

L41 ANSWER 36 OF 92 EMBASE COPYRIGHT 2001 ELSEVIER SCI. B.V.
AN 1998251081 EMBASE
TI Influence of tyrosine and phenylalanine limitation on cytotoxicity of ***chimeric*** TGF-.alpha. toxins on B16BL6 murine melanoma In vitro.
AU Fu Y.-M.; Li Y.-Q.; Meadows G.G.
CS Dr. G.G. Meadows, Dept. of Pharmaceutical Sciences, Box 646510, College of Pharmacy, Pullman, WA 99164-6510, United States. meadows@mail.wsu.edu
SO Nutrition and Cancer, (1998) 31/1 (1-7).
Refs: 31
ISSN: 0163-5581 CODEN: NUCADQ
CY United States
DT Journal; Article
FS 016 Cancer
029 Clinical Biochemistry
LA English
SL English
AB Previous research in animals supports the use of tyrosine and phenylalanine (Tyr-Phe) restriction as an adjuvant to the treatment of cancer. In this regard, dietary restriction of Tyr-Phe specifically inhibits the growth of B16BL6 melanoma tumors, dramatically suppresses spontaneous hematogenous metastasis, and modulates the sensitivity of these tumor cells to growth factors. Two ***chimeric*** toxins, HB-TGF.alpha.-PE(4E) ***KDEL*** and TGF.alpha.- PE(4E) ***KDEL*** , were examined for their toxicity against the B16BL6 melanoma cell line, and the ability of Tyr-Phe limitation to modulate the potential of these toxins was examined. Tyr-Phe limitation significantly enhanced the cytotoxic effects of HBTGF.alpha.-pE(4E) ***KDEL*** approximately 10-fold toward B16BL6 melanoma, and free heparin diminished the cytotoxicity of HB-TGF.alpha.- PE(4E) ***KDEL*** . Although TGF.alpha.-PE(4E) ***KDEL*** is cytotoxic to this cell line, Tyr- Phe limitation did not affect the cytotoxicity of this toxin. Tyr-Phe limitation inhibited the synthesis and secretion of heparin-binding proteins but did not alter the expression of surface heparan sulfate proteoglycans. These data suggest that cell surface heparan sulfate proteoglycan is a target for binding and execution of the cytotoxicity of HB-TGF.alpha.-PE(4E) ***KDEL*** and that augmentation of cytotoxicity by Tyr-Phe limitation is due to the inhibition of heparin-binding protein production.

L41 ANSWER 37 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 24
AN 1998:44502 BIOSIS
DN PREV199800044502
TI The mammalian protein (rbet1) homologous to yeast Bet1p is primarily associated with the pre-Golgi intermediate compartment and is involved in vesicular transport from the endoplasmic reticulum to the Golgi apparatus.
AU Zhang, Tao; Wong, Siew Heng; Tang, Bor Luen; Xu, Yue; Peter, Frank; Subramaniam, V. Nathan; Hong, Wanjin (1)
CS (1) Membrane Biol. Lab., Inst. Molecular Cell Biol., 15 Lower Kent Ridge Rd., Singapore 119076 Singapore
SO Journal of Cell Biology, (Dec. 1, 1997) Vol. 139, No. 5, pp. 1157-1168.
ISSN: 0021-9525.
DT Article
LA English
AB Yeast Bet1p participates in vesicular transport from the endoplasmic reticulum to the Golgi apparatus and functions as a soluble N-ethylmaleimide-sensitive factor attachment protein receptor (SNARE) associated with ER-derived vesicles. A mammalian protein (rbet1) homologous to Bet1p was recently identified, and it was concluded that rbet1 is associated with the Golgi apparatus based on the subcellular localization of transiently expressed epitope-tagged rbet1. In the present study using rabbit antibodies raised against the cytoplasmic domain of rbet1, we found that the majority of rbet1 is not associated with the Golgi apparatus as marked by the Golgi mannosidase II in normal rat kidney cells. Rather, rbet1 is predominantly associated with vesicular spotty structures that concentrate in the peri-Golgi region but are also present throughout the cytoplasm. These structures colocalize with the ***KDEL*** receptor and ERGIC-53, which are known to be enriched in the intermediate compartment. When the Golgi apparatus is fragmented by nocodazole treatment, a significant portion of rbet1 is not colocalized with structures marked by Golgi mannosidase II or the ***KDEL*** receptor. Association of rbet1 in cytoplasmic spotty structures is apparently not altered by preincubation of cells at 15degreeC. However, upon warming up from 15 to 37degreeC, rbet1 concentrates into the peri-Golgi region. Furthermore, rbet1 colocalizes with vesicular stomatitis virus G-protein en route from the ER to the Golgi. Antibodies against rbet1 inhibit in vitro transport of G-protein from the ER to the Golgi apparatus in a dose-dependent manner. This inhibition can be neutralized by preincubation of antibodies with recombinant rbet1. EGTA is known to inhibit ER-Golgi transport at a stage after vesicle docking but before the actual ***fusion*** event. Antibodies against rbet1 inhibit ER-Golgi transport only when they are added before the EGTA-sensitive stage. These results suggest that rbet1 may be involved in the docking process of ER-derived vesicles with the cis-Golgi membrane.

L41 ANSWER 38 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 25
AN 1997:244348 BIOSIS
DN PREV199799543551
TI Dissociation of coatomer from membranes is required for brefeldin A-induced transfer of golgi enzymes to the endoplasmic reticulum.
AU Scheel, Jochen; Pepperkok, Rainer; Lowe, Martin; Griffiths, Gareth; Kreis, Thomas E. (1)
CS (1) Dep. Cell Biol., Sciences III, Univ. Geneva, Geneva Switzerland
SO Journal of Cell Biology, (1997) Vol. 137, No. 2, pp. 319-333.
ISSN: 0021-9525.
DT Article
LA English
AB Addition of brefeldin A (BFA) to mammalian cells rapidly results in the removal of coatomer from membranes and subsequent delivery of Golgi enzymes to the endoplasmic reticulum (ER). Microinjected anti-EAGE (intact IgG or Fab-fragments), antibodies against the "EAGE"-peptide of beta-COP, inhibit BFA-induced redistribution of beta-COP in vivo and block transfer of resident proteins of the Golgi complex to the ER; tubulo-vesicular clusters accumulate and Golgi membrane proteins concentrate in cytoplasmic patches containing beta-COP. These patches are devoid of marker proteins of the ER, the intermediate compartment (IC), and do not contain ***KDEL*** receptor. Interestingly, relocation of ***KDEL*** receptor to the IC, where it colocalizes with ERGIC53 and ts-O45-G, is not inhibited under these conditions. While no stacked Golgi cisternae remain in these injected cells, reassembly of stacks of Golgi cisternae following BFA wash-out is inhibited to only apprx 50%. Mono- or divalent anti-EAGE stabilize binding of coatomer to membranes in vitro, at least as efficiently as GTP-gamma-S. Taken together these results suggest that enhanced binding of coatomer to membranes completely inhibits the BFA-induced retrograde transport of Golgi resident proteins to the ER, probably by inhibiting ***fusion*** of Golgi with ER membranes, but does not interfere with the disassembly of the stacked Golgi cisternae and recycling of ***KDEL*** receptor to the IC. These results confirm our previous results suggesting that COPI is involved in anterograde membrane transport from the ER/IC to the Golgi complex (Pepperkok et al., 1993), and corroborate that COPI regulates retrograde membrane transport between the Golgi complex and ER in mammalian cells.

L41 ANSWER 39 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 26
AN 1997:163727 BIOSIS
DN PREV199799462930
TI The C-terminal HDEL sequence is sufficient for retention of secretory proteins in the endoplasmic reticulum (ER) but promotes vacuolar targeting of proteins that escape the ER.
AU Gomord, Veronique; Denmat, Lise-Anne; Fitchette-Laine, Anne-Catherine; Satiat-Jeunemaitre, Beatrice; Hawes, Chris; Faye, Loic (1)
CS (1) LTI-CNRS URA 203, UFR Sci., IFRMP 23, Univ. Rouen, 76821 Mt. St. Aignan Cedex France
SO Plant Journal, (1997) Vol. 11, No. 2, pp. 313-325.
ISSN: 0960-7412.
DT Article
LA English
AB Proteins are co-translationally transferred into the endoplasmic reticulum (ER) and then either retained or transported to different intracellular compartments or to the extracellular space. Various molecular signals necessary for retention in the ER or targeting to different compartments have been identified. In particular, the HDEL and ***KDEL*** signals used for retention of proteins in yeast an animal ER have also been described at the C-terminal end of soluble ER processing enzymes in plants. The ***fusion*** of a ***KDEL*** extension to vacuolar proteins is sufficient for their retention in the ER of transgenic plant cells. However, recent results obtained using the same strategy indicate that HDEL does not contain sufficient information for full retention of phaseolin expressed in tobacco. In the present study, an HDEL C-terminal extension was fused to the vacuolar or extracellular (DELTA-pro) forms of sporamin. The resulting SpoHDEL or DELTA-proHDEL, as well as Spo and DELTA-pro, were expressed at high levels in transgenic tobacco cells (Nicotiana tabacum cv BY2). The intracellular location of these different forms of recombinant sporamin was studied by subcellular fractionation. The results clearly indicate that addition of an HDEL extension to either Spo or DELTA-pro induces accumulation of these sporamin forms in a compartment that co-purifies with the ER markers NADH cytochrome C reductase, binding protein (BiP) and calnexin. In addition, a significant SpoHDEL or DELTA-proHDEL fraction that escapes the ER retention machinery is transported to the vacuole. From these results, it may be proposed that, in addition to its function as an ER retention signal, HDEL could also act in quality control by targeting chaperones or chaperone-bound proteins that escape the ER to the plant lysosomal compartment for degradation.

L41 ANSWER 40 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 27
AN 1997:201060 BIOSIS
DN PREV199799500263
TI Nuclear localisation of calreticulin in vivo is enhanced by its interaction with glucocorticoid receptors.
AU Roderick, H. Llewelyn; Campbell, Anthony K.; Llewellyn, David H. (1)
CS (1) Dep. Med. Biochem., Univ. Wales Coll. Med., Heath Park, Cardiff CF4 4XN UK
SO FEBS Letters, (1997) Vol. 405, No. 2, pp. 181-185.
ISSN: 0014-5793.

DT Article
LA English
AB The multi-functional protein calreticulin (CRT) is normally found within the lumen of the endoplasmic reticulum (ER). However, some of its proposed functions require it to be located within the nucleus, where its presence is contentious. We have investigated this in live COS7, HeLa and LM(TK-) cells using green fluorescent protein (GFP)- ***fusion*** proteins. GFP-CRT, and GFP, with an ER signal peptide and a ***KDEL*** sequence (ER-GFP), were localized to the ER. In addition, GFP-CRT was located in the nucleus of all the cell types at low levels. The higher levels of nuclear fluorescence in LM(TK-) and HeLa cells suggested that glucocorticoid receptors might enhance nuclear localization of calreticulin. Dexamethasone treatment of LM(TK-) cells doubled the amount of nuclear GFP-CRT, but did not affect the localization of a GFP-CRT ***fusion*** in which the glucocorticoid receptor-binding N-domain of calreticulin had been deleted. Thus, despite ER targeting and retention signals, calreticulin is also located within the nucleus where its presence increases due to its interaction with glucocorticoid receptors.

L41 ANSWER 41 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 28
AN 1997:152908 BIOSIS
DN PREV199799452111
TI Characterization of brefeldin A induced vesicular structures containing cycling proteins of the intermediate compartment/cis-Golgi network.
AU Fuellekrug, Joachim; Soennichsen, Birte; Schaefer, Ulrike; Van, Phuc Nguyen; Soeling, Hans-Dieter; Mieskes, Gottfried (1)
CS (1) Dep. Clinical Biochemistry, Univ. Goettingen, Robert-Koch-Str. 40, D-37075 Goettingen Germany
SO FEBS Letters, (1997) Vol. 404, No. 1, pp. 75-81.
ISSN: 0014-5793.
DT Article
LA English
AB Residence of luminal ER proteins is mediated by a cyclic process which involves binding of escaped proteins to a ***KDEL*** receptor in a post-ER compartment and redistribution of the ligand-receptor complex back to the ER. We examined the relocation of the ***KDEL*** receptor after treatment with the fungal metabolite brefeldin A and compared this with the retrograde transport of the ***KDEL*** receptor observed after ligand or receptor overexpression. Incubation with brefeldin A led to the formation of vesicular structures containing the ***KDEL*** receptor and ERGIC-53, a marker for the ER-Golgi intermediate compartment. Immunoelectron microscopy revealed that these structures induced vesicular structures were morphologically and biochemically distinct from the ER-Golgi ***hybrid*** compartment as subcellular fractionation. Overexpression of the receptor itself or together with ERGIC-53, an intermediate compartment marker to the ER but not to structures resembling BFA induced vesicular structures. Moreover, overexpression of the receptor resulted in the partial redistribution of marker proteins of the medial Golgi and the trans-Golgi network to ER-like structures. We conclude that the effects of brefeldin A on the redistribution of the ***KDEL*** receptor do not reflect physiological events occurring during increased occupancy of the receptor with ligands.

L41 ANSWER 42 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 29
AN 1996:408998 BIOSIS
DN PREV199699131354
TI Interactions between microsomal triglyceride transfer protein and apolipoprotein B within the endoplasmic reticulum in a ***heterologous*** expression system.
AU Patel, Shailendra B. (1); Grundy, Scott M.
CS (1) Dep. Intern. Med., Univ. Texas Southwestern Med. Cent., Y3.208, 5323 Harry Hines Blvd., Dallas, TX 75235-9052 USA
SO Journal of Biological Chemistry, (1996) Vol. 271, No. 31, pp. 18686-18694.
ISSN: 0021-9258.
DT Article
LA English
AB When apolipoprotein B (apoB) is expressed in ***heterologous*** cells, it is not secreted but retained and degraded within the endoplasmic reticulum (ER). We have previously characterized carboxyl-terminal truncated forms of apoB expressed in COS cells and have shown that these proteins were readily synthesized but retained within the ER and degraded, if the size of the truncated protein was larger than apoB 29. Below this size, the smaller the size of the apoB truncates, the greater the extent of secretion, although gt 50% of these smaller proteins were also degraded within the ER. In the present study, we demonstrate that this secretory defect can be overcome by coexpression with microsomal triglyceride transfer protein (MTP); moreover, this complementation is inversely related to the size of apoB. Secretion of apoBs larger than B29 required the coexpression of MTP and, in the presence of MTP, was oleate-responsive. MTP, in the presence or absence of oleate supplementation, had little or no effect on the secretion of the shorter truncates. We discovered, however, that MTP was physically associated with all forms of apoB intracellularly (B13-B41). The association of MTP with apoB 41 was stable to high salt washing, as well as to low pH, suggesting that these interactions may be hydrophobic in nature. In addition to the interaction with MTP, apoB was also found to be associated with calnexin, confirming previous studies, and with proteins bearing the ***KDEL*** retention signal. However, studies on overexpression of human calnexin and tunicamycin inhibition of glycosylation showed that interaction with calnexin was not necessary for the formation or secretion of apoB 41-containing lipoproteins; moreover, in the presence of MTP, the association of calnexin with apoB 41 was transient or absent. These data suggest that for apoB to attain a folded state sufficient to escape the quality control of the ER, it needs to obtain neutral lipid (supplied by MTP), as well as its ability to keep it packaged as a rudimentary lipoprotein, dependent on its size being larger than B29.

L41 ANSWER 43 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 30
AN 1996:411789 BIOSIS
DN PREV199699134145
TI Down-regulation of paramyxovirus hemagglutinin-neuraminidase glycoprotein surface expression by a mutant ***fusion*** protein containing a retention signal for the endoplasmic reticulum.
AU Tanaka, Yoshikazu; Heminway, Beverly R.; Galinski, Mark S. (1)
CS (1) Merck Co. Inc., P.O. Box 4, WP29M-4, Sumneytown Pike, West Point, PA 19486 USA
SO Journal of Virology, (1996) Vol. 70, No. 8, pp. 5005-5015.
ISSN: 0022-538X.
DT Article
LA English
AB The human parainfluenza virus type 3 (HPIV3) ***fusion*** (F) and hemagglutinin-neuraminidase (RN) glycoproteins are the principal components involved in virion receptor binding, membrane penetration, and ultimately, syncytium formation. While the requirement for both F and RN in this process has been determined from recombinant expression studies, stable physical association of these proteins in coimmunoprecipitation studies has not been observed. In addition, coexpression of other ***heterologous*** paramyxovirus F or RN glycoproteins with either HPIV3 F or RN does not result in the formation of syncytia, suggesting serotypespecific protein differences. In this study, we report that simian virus 5 and Sendai virus ***heterologous*** RN proteins and measles virus hemagglutinin (H) were found to be down-regulated when coexpressed with HPIV3 F. As an alternative to detecting physical associations of these proteins by coimmunoprecipitation, further studies were performed with a mutant HPIV3 F protein (F- ***KDEL***) lacking a transmembrane anchor and cytoplasmic tail and containing a carboxyl-terminal retention signal for the endoplasmic reticulum (ER). F- ***KDEL*** was defective for transport to the cell surface and could down-regulate surface expression of HPIV3 RN and ***heterologous*** HN/H proteins from simian virus 5, Sendai virus, and measles virus in coexpression experiments. HN/H down-regulation appeared to result, in part, from an early block to HPIV3 RN synthesis, as well as an instability of the ***heterologous*** HN/H proteins within the ER. In contrast, coexpression of F- ***KDEL*** with HPIV3 wild-type F or the ***heterologous*** receptor-binding proteins, respiratory syncytial virus glycoprotein (G) and vesicular stomatitis virus glycoprotein (G), were not affected in transport to the cell surface. Together, these results support the notion that the reported serotype-specific restriction of syncytium formation may involve, in part, down-regulation of ***heterologous*** HN expression.

L41 ANSWER 44 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 31
AN 1996:181995 BIOSIS
DN PREV199698738124
TI Recombinant immunotoxin containing a disulfide-stabilized Fv directed at erbB2 that does not require proteolytic activation.
AU Kuan, Chien-Tsun; Pastan, Ira (1)
CS (1) Lab. Mol. Biol., Div. Basic Sci., Natl. Cancer Inst., NIH, Build. 37, Room 4E16, 37 Convent Drive, MSC 4255, Bethesda, MD 20892-4255 USA
SO Biochemistry, (1996) Vol. 35, No. 9, pp. 2872-2877.
ISSN: 0006-2960.
DT Article
LA English
AB PE35/e23(dsFv) ***KDEL*** is a recombinant immunotoxin composed of a recombinant form of Pseudomonas exotoxin that does not need proteolytic activation and a disulfide-stabilized Fv fragment of the anti-erbB2 monoclonal antibody e23. In this molecule, the variable heavy (V-H) domain is inserted near the carboxyl terminus of PE at position 607 and the variable light (V-L) domain is connected to the VH domain by a disulfide bond engineered into the framework region. The disulfide bond forms between cysteines introduced at position 44 Of VH and position 99 of V-L (Reiter et al. (1994) J. Biol. Chem. 269, 18327-18331). In contrast to other PE-defined Fv ***fusion*** proteins, this type of recombinant toxin does not need proteolytic activation of the toxin domain. PE35/e23(dsFv) ***KDEL*** is very cytotoxic toward erbB2 antigen-expressing N87 cells (IC-50 = 0.8 ng/mL) despite the fact that it binds to the erbB2 protein only 25% as well as e23(dsFv)PE38KDEL, in which the dsFv moiety is located at the amino terminus of the toxin. The lower binding affinity is probably due to interference by domain III of PE with the amino terminus of e23(V-H), possibly where the antigen binding sites are located. Nevertheless, the specificity of immunotoxin is still retained, and it is very stable at 37 degree C. Because of its small size, stability, and activity without proteolytic processing, this immunotoxin may be advantageous for tumor treatment. PE35/ e23(dsFv) ***KDEL*** was also used to gain information about whether reduction of the disulfide bonds connecting V-H and V-L occur in the endoplasmic reticulum (ER) or in a proximal compartment. To do this, we switched the ER retention sequence ***KDEL*** from the toxin-V-H subunit to the V-L subunit. Our results suggest that reduction of the disulfide bond connecting the dsFv heterodimer occurs before the immunotoxin reaches the ER, where translocation to the cytosol appears to occur.

L41 ANSWER 45 OF 92 CAPLUS COPYRIGHT 2001 ACS
AN 1997:1462 CAPLUS
DN 126:85202
TI Virus-mediated delivery of the green fluorescent protein to the

endoplasmic reticulum of plant cells
AU Boevink, P.; Santa Cruz, S.; Hawes, C.; Harris, N.; Oparka, K. J.
CS Dep. Cellular Environmental Physiology, Scottish Crop Research Inst., Invergowrie, Dundee, DD2 5DA, UK
SO Plant J. (1996), 10(5), 935-941
CODEN: PLJUED; ISSN: 0960-7412
PB Blackwell
DT Journal
LA English
AB The green fluorescent protein (GFP)from Aequorea victoria was targeted to the endoplasmic reticulum (ER) of living plant cells using a virus-based expression system. The signal peptide from the storage protein, patatin was fused to the N-terminus of the GFP, whereas the ER retention signal ***KDEL*** was fused to the C-terminus. The ***chimeric*** gfp cDNA was inserted into a potato virus X-based expression vector and in-vitro transcripts, representing the recombinant viral genome, were inoculated on to Nicotiana benthamiana and N. clevelandii plants. In virus-infected cells, the GFP was targeted both to the cortical ER and to a mobile system of ER elements which underwent streaming in the cytoplasm. In addn., a population of GFP-contg. inclusions was apparent. These inclusions were motile but remained closely assocd. with elements of the ER. Staining of the ER with membrane potential-sensitive dyes confirmed that the GFP had been targeted to the ER. The utility of virus-mediated delivery systems in studies of the plant endomembrane system is discussed.

L41 ANSWER 46 OF 92 EMBASE COPYRIGHT 2001 ELSEVIER SCI. B.V.DUPLICATE 32
AN 96187449 EMBASE
DN 1996187449
TI Bip/GRP78 but not calnexin associates with a precursor of glycosylphosphatidylinositol-anchored protein.
AU Oda K.; Wada I.; Takami N.; Fujiwara T.; Misumi Y.; Ikehara Y.
CS Department of Biochemistry, Niigata University School Dentistry,Niigata 951, Japan
SO Biochemical Journal, (1996) 316/2 (623-630).
ISSN: 0264-6021 CODEN: BIJOAK
CY United Kingdom
DT Journal; Article
FS 029 Clinical Biochemistry
LA English
SL English
AB When fused in-frame with a C-terminal propeptide of placental alkaline phosphatase (PLAP), rat .alpha.(2u)-globulin (.alpha.GL), a nonglycosylated secretory protein, was expressed on the cell surface as a glycosylphosphatidylinositol (GPI)-linked chimaeric protein (.alpha.GL-PLAP). In contrast with the wild-type .alpha.GL-PLAP, a mutant, in which Asp at the cleavage/attachment site of GPI was replaced by Trp, failed to become a GPI-linked mature form and was retained as a precursor form within the cell. To elucidate the molecular interactions involved in the retention of the preform within the cell, we examined the association of the preform with molecular chaperones in the endoplasmic reticulum (ER). Antibody against the ER retrieval motif ***KDEL*** coimmunoprecipitated a 25 kDa preform, but not a 22 kDa GPI-linked mature form. Pulse-chase experiments showed that the wild-type .alpha.GL-PLAP with a cleavable propeptide was converted into the mature form, whereas the mutant .alpha.GL-PLAP with an uncleavable propeptide remained associated with ER-resident proteins with a ***KDEL*** motif and underwent rapid degradation in a pre-Golgi compartment. Chemical cross-linking studies showed that, of the several ER-resident proteins immunoreactive with the anti- ***KDEL*** antibody, a 78 kDa protein was the only protein associated with the preform. Furthermore this 78 kDa protein was dissociated from the precursor molecule on incubation with ATP, allowing us tentatively to assign it as Bip/GRP78. Anti-calnexin antibody, however, failed to coprecipitate any form of the chimaeric protein. Immunoelectron microscopy showed that the preform with the uncleavable propeptide was localized in the ER, but not detected in the Golgi apparatus or plasma membranes. Taken together, these results suggest that Bip/GRP78 is associated with pro.alpha.GL-PLAP and retains it within the ER until pro.alpha.GL-PLAP is either modified by GPI or degraded, thereby participating in the quality control of this GPI-linked chimaeric protein.

L41 ANSWER 47 OF 92 CAPLUS COPYRIGHT 2001 ACS
AN 1996:760118 CAPLUS
DN 126:30254
TI Replication of primary HIV-1 isolates is inhibited in PM1 cells expressing sCD4- ***KDEL***
AU Degar, Steven; Johnson, J. Erik; Boritz, Eli; Rose, John K.
CS Dep. Pathology Cell Biology, Yale Univ. Sch. Med., New Haven, CT, 06510, USA
SO Virology (1996), 226(2), 424-429
CODEN: VIRLAX; ISSN: 0042-6822
PB Academic
DT Journal
LA English
AB Expression of a sol. CD4 mol. (sCD4- ***KDEL***) contg. a specific retention signal for the endoplasmic reticulum was shown previously to block propagation of the HIV-1MN prototype strain in a transformed T cell line. However, the virus present in HIV-1-infected individuals is more closely represented by primary HIV-1 isolates which, unlike the HIV-1MN strain, have not been adapted to growth in cell lines. To det. if sCD4- ***KDEL*** could block replication of primary isolates the authors used the PM1 cell line that has been shown to propagate primary isolates without adaptation. Here the authors show that the replication of four primary HIV-1 isolates was strongly inhibited in PM1 cells that expressed sCD4- ***KDEL*** under control of the HIV-1 LTR. Infection with primary HIV-1 isolates induced sCD4- ***KDEL*** expression driven by the LTR, HIV-1 spread was dramatically reduced, and reverse transcriptase activity in the cell culture supernatants was greatly diminished. SCD4- ***KDEL*** , therefore, represents a potent inhibitor of HIV-1 replication for gene therapy-based approaches for the treatment of AIDS.

L41 ANSWER 48 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 33
AN 1997:107341 BIOSIS
DN PREV199799406544
TI The dynamic organisation of the secretory pathway.
AU Pelham, Hugh R. B.
CS MRC Lab. Molecular Biol., Hills Rd., Cambridge CB2 2QH UK
SO Cell Structure and Function, (1996) Vol. 21, No. 5, pp. 413-419.
ISSN: 0386-7196.
DT General Review
LA English
AB The secretory pathway of eukaryotic cells consists of a number of distinct membrane-bound compartments interconnected by vesicular traffic. Each compartment has a characteristic content of proteins and lipids, which must be maintained. This is achieved in most cases by active sorting - proteins may reach the wrong compartment but are continually retrieved. A good example is the retrieval system for lumenal ER proteins. These proteins carry a specific sorting signal, typically the tetrapeptide ***KDEL*** , which is bound by a receptor in the Golgi apparatus. The receptor-ligand complex, together with escaped ER membrane proteins, returns to the ER. Many of the components of vesicle traffic, including the coat proteins required for vesicle budding from the ER, those that form retrograde vesicles on post-ER compartments, and integral membrane proteins that target the vesicles to their correct destination, have been identified. The sorting events that occur can largely be understood in terms of specific protein-protein interactions involving these components. However, sorting of some membrane proteins, including the vesicle targeting molecules, is influenced by their transmembrane domains, and it is likely that segregation of these is dependent on the composition and biophysical properties of the lipid bilayer, which very between compartments. The secretory pathway is thus a dynamic entity, split into discrete organelles by the constant segregation and recycling of lipids and proteins, processes that are ultimately driven by the mechanics of vesicle formation and ***fusion*** .

L41 ANSWER 49 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 34
AN 1996:576655 BIOSIS
DN PREV199799291336
TI Structural change of the endoplasmic reticulum during fertilization: Evidence for loss of membrane continuity using the green fluorescent protein.
AU Terasaki, Mark (1); Jaffe, Laurinda A.; Hunnicutt, Gary R.; Hammer, John A., III
CS (1) Marine Biol. Lab., Woods Hole, MA 02543 USA
SO Developmental Biology, (1996) Vol. 179, No. 2, pp. 320-328.
ISSN: 0012-1606.
DT Article
LA English
AB Green fluorescent protein (GFP) was targeted to the lumen of the endoplasmic reticulum (ER) of starfish eggs by injecting mRNA coding for a ***chimeric*** protein containing a signal sequence and the ***KDEL*** ER retention sequence. By confocal microscopy, the GFP ***chimeric*** protein was localized in intracellular cisternae (membrane sheets) and the nuclear envelope, showing that it had been successfully targeted to the ER. The labeling pattern closely resembled that produced by the fluorescent dicarbocyanine DiI, which has been used previously to label the ER (Jaffe and Terasaki, Dev. Biol. 164, 579-587, 1994). Eggs expressing the GFP chimera were used to examine whether there is a loss of ER continuity at fertilization. The time required for recovery of fluorescence after photobleaching for both the GFP chimera and DiI was much longer in eggs at 1 min postfertilization than in unfertilized eggs or in 20-min-postfertilized eggs. This result provides strong evidence for a transient loss of continuity of the ER associated with Ca release at fertilization.

L41 ANSWER 50 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 35
AN 1996:120026 BIOSIS
DN PREV199698692161
TI Cytotoxicity of ***KDEL*** -terminated ricin toxins correlates with distribution of the ***KDEL*** receptor in the Golgi.
AU Tagge, Edward; Chandler, John; Tang, Bor Luen; Hong, Wanjin; Willingham, Mark C.; Frankel, Arthur (1)
CS (1) Hollings Cancer Cent., Room 306, Medical U. South Carolina, 171 Ashley Ave., Charleston, SC 29425 USA
SO Journal of Histochemistry and Cytochemistry, (1996) Vol. 44, No. 2, pp. 159-165.
ISSN: 0022-1554.
DT Article
LA English
AB DNAs encoding ricin toxin A chain (RTA), with or without a C-terminal endoplasmic reticulum retention signal ***KDEL*** , were subcloned into pGEX2T bacterial expression plasmid. After transformation of JM105 E. coli cells and induction with isopropylthio-beta-galactoside (IPTG), ***fusion*** proteins were bound to an immobilized glutathione matrix and recombinant ricin A chains released with thrombin. Both recombinant

wild-type RTA and RTA with ***KDEL*** had immunological reactivity and catalytic activity indistinguishable from plant RTA. The bacterial RTA products reassociated with plant ricin B chain (RTB) similarly to plant RTA. Cell cytotoxicities were measured on seven cell lines for each A-chain and heterodimer. Although ***KDEL*** sequences enhanced cytotoxicity in most cases, significant variability was observed. In each case, addition of ***KDEL*** enhanced A-chain cytotoxicity more than holotoxin cytotoxicity. Three cell lines showed reduced ***KDEL*** enhancement of both RTA and ricin cytotoxicity. The concentration of ***KDEL*** receptor was examined on each cell line by immunofluorescence microscopy with an antireceptor monoclonal antibody. Differences in sensitivity to ***KDEL*** -containing toxins correlated with altered distribution of ***KDEL*** receptor between endoplasmic reticulum (ER) and Golgi compartments.

L41 ANSWER 51 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 36
AN 1996:187769 BIOSIS
DN PREV199698743898
TI Molecular characterization of cDNAs encoding low-molecular-weight heat shock proteins of soybean.
AU Lafayette, Peter R.; Nagao, Ronald T. (1); O'Grady, Kevin; Vierling, Elizabeth; Key, Joe L.
CS (1) Department Botany, University Georgia, Athens, GA 30602 USA
SO Plant Molecular Biology, (1996) Vol. 30, No. 1, pp. 159-169.
ISSN: 0167-4412.
DT Article
LA English
AB Three cDNA clones (GmHSP23.9, GmHSP22.3, and GmHSP22.5) representing three different members of the low-molecular-weight (LMW) heat shock protein (HSP) gene superfamily were isolated and characterized. A fourth cDNA clone, pFS2033, was partially characterized previously as a full-length genomic clone GmHSP22.0. The deduced amino acid sequences of all four cDNA clones have the conserved carboxyl-terminal LMW HSP domain. Sequence and hydropathy analyses of GmHSP22, GmHSP22.3, and GmHSP22.5, representing HSPs in the 20 to 24 kDa range, indicate they contain amino-terminal signal peptides. The mRNAs from GmHSP22, GmHSP22.3, and GmHSP22.5 were preferentially associated in vivo with endoplasmic reticulum (ER)-bound polysomes. GmHSP22 and GmHSP22.5 encode strikingly similar proteins; they are 78% identical and 90% conserved at the amino acid sequence level, and both possess the C-terminal tetrapeptide KQEL which is similar to the consensus ER retention motif ***KDEL*** ; the encoded polypeptides can be clearly resolved from each other by two-dimensional gel analysis of their ***hybrid*** -arrest translation products. GmHSP22.3 is less closely related to GmHSP22 (48% identical and 70% conserved) and GmHSP22.5 (47% identical and 65% conserved). The fourth cDNA clone, GmHSP23.9, encodes a HSP of ca. 24 kDa with an amino terminus that has characteristics of some mitochondrial transit sequences, and in contrast to GmHSP22, GmHSP22.3, and GmHSP22.5, the corresponding mRNA is preferentially associated in vivo with free polysomes. It is proposed that the LMW HSP gene superfamily be expanded to at least six classes to include a mitochondrial class and an additional endomembrane class of LMW HSPs.

L41 ANSWER 52 OF 92 EMBASE COPYRIGHT 2001 ELSEVIER SCI. B.V.
AN 97040775 EMBASE
DN 1997040775
TI Cell killing effect of heparin-binding EGF-like growth factor Pseudomonas exotoxin on human hepatoma cells.
AU Ono M.; Klagsbrun M.; Kohgo Y.
CS M. Ono, Third Dept. of Internal Medicine, Asahikawa Medical College, 4-5 Nishikagura, Asahikawa, Hokkaido 078, Japan
SO International Hepatology Communications, (1996) 6/2 (79-84).
Refs: 12
ISSN: 0928-4346 CODEN: IHCOEP
PUI S 0928-4346(96)00333-7
CY Ireland
DT Journal; Article
FS 029 Clinical Biochemistry
048 Gastroenterology
037 Drug Literature Index
LA English
SL English
AB Heparin-binding, EGF-like growth factor (HB-EGF) is a potent mitogen for smooth muscle cell, fibroblast, and it also stimulates hepatocyte proliferation. We generated several ***chimeric*** toxins by fusing the cDNA sequence of HB-EGF and the mutant of Pseudomonas exotoxin, PE(4E) ***KDEL*** (PE) that lacks the binding ability to a specific receptor. HB-EGF-PE was generated by fusing the DNA fragment encoding the full length mature HB-EGF polypeptide to the N-terminus of PE(4E) ***KDEL*** , while HB-PE was generated by fusing the 45 N-terminal heparin-binding sequence to PE(4E) ***KDEL*** . HB-EGF-PE was capable of binding both to the EGF receptor and heparin sulfate proteoglycans (HSPGs), whereas HB-PE was capable of binding only to HSPGs on the target cells. Human hepatoma cells, SK-Hep1, Hep-G2 and PLC/PRF/5 were killed in a very low concentration, of HB-EGF-PE with the ID50 of 0.1-0.5 ng/ml. HB-PE could also kill SK-Hep1 with the ID50 of 50 ng/ml, whereas it was resistant to PE. Both exogenous EGF and heparin inhibited the cytotoxicity of HB-EGF-PE. These results indicated the existence of two alternative pathways for the internalization of the ***chimeric*** toxins defined by two different targets, EGFR and HSPGs.

L41 ANSWER 53 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 37
AN 1996:289304 BIOSIS
DN PREV199699011660
TI Enhancement of cytotoxic activity of interleukin 2-pseudomonas ***fusion*** proteins.
AU Gao Jimin, Xu Lingfei (1); Zheng Zhongcheng; et al.
CS (1) First Military Med. Univ., Guangzhou 510515 China
SO Zhonghua Weishengwuxue He Mianyixue Zazhi, (1996) Vol. 16, No. 1, pp. 37-40.
ISSN: 0254-5101.
DT Article
LA Chinese
SL Chinese; English
AB We confirmed that the IL2-PE ***fusion*** proteins such as IL2-PE40REDLK, IL2PE4oKDEL,IL2-PE66-4GluREDLK and IL2-PE66-4GluKDEL had potent cytotoxicity on the target cells with high affinity IL-2R through the specific binding of IL-2 moiety to IL-2R and the toxicity of the PE moiety. The results showed that the sensitivity of the same target cells to various IL2-PE ***fusion*** proteins were different (for example,the cytotoxicity of IL2-PE66-4GluKDEL on the PHA-activated blasts of human peripheral blood was about 40 times as much as that of IL2-PE40REDLK). It was also discovered that the carbonyl terminal REDLK sequence of IL-2-PE ***fusion*** proteins substituted by ***KDEL*** would enhance their cytotoxicity (for example, the cytotoxicity of IL2-PE40 ***KDEL*** on the ConA-stimulated murine spleen blasts was about 8 times than the one of IL2-PE40REDLK).

L41 ANSWER 54 OF 92 EMBASE COPYRIGHT 2001 ELSEVIER SCI. B.V.
AN 96381971 EMBASE
DN 1996381971
TI Signal-mediated sorting of membrane proteins between the endoplasmic reticulum and the Golgi apparatus.
AU Teasdale R.D.; Jackson M.R.
CS R. W. Johnson Pharmaceut. Res. Inst., 3535 General Atomics Court,San Diego, CA 92121, United States
SO Annual Review of Cell and Developmental Biology, (1996) 12/- (27-54).
ISSN: 1081-0706 CODEN: ARDBF8
CY United States
DT Journal; General Review
FS 029 Clinical Biochemistry
LA English
SL English
AB Each organelle of the secretory pathway is required to selectively allow transit of newly synthesized secretory and plasma membrane proteins and also to maintain a unique set of resident proteins that define its structural and functional properties. In the case of the endoplasmic reticulum (ER), residency is achieved in two ways: (a) prevention of residents from entering newly forming transport vesicles and (b) retrieval of those residents that escape. The latter mechanism is directed by discrete retrieval motifs: Soluble proteins have a H/ ***KDEL*** sequence at their carboxy-terminus; membrane proteins have a dibasic motif, either di-lysine or di-arginine, located close to the terminus of their cytoplasmic domain. Recently it was found that di- lysine motifs bind the complex of cytosolic coat proteins, COP I, and that this interaction functions in the retrieval of proteins from the Golgi to the ER. Also discussed are the potential roles this interaction may have in vesicular trafficking.

L41 ANSWER 55 OF 92 EMBASE COPYRIGHT 2001 ELSEVIER SCI. B.V.
AN 95229992 EMBASE
DN 1995229992
TI Increased antitumor activity of a circularly permuted interleukin 4-toxin in mice with interleukin 4 receptor-bearing human carcinoma.
AU Kreitman R.J.; Puri R.K.; Pastan I.
CS Laboratory of Molecular Biology, National Cancer Institute/NIH, Building 37, 37 Convent Drive,Bethesda, MD 20892-4255, United States
SO Cancer Research, (1995) 55/15 (3357-3363).
ISSN: 0008-5472 CODEN: CNREA8
CY United States
DT Journal; Article
FS 016 Cancer
026 Immunology, Serology and Transplantation
037 Drug Literature Index
LA English
SL English
AB We reported previously that circularly permuted interleukin-4 (IL4), composed of amino acids 38-129 of IL4 connected by a linker peptide GGNGG to amino acids 1-37, is preferable to native IL4 for fusing to the amino terminus of truncated Pseudomonas exotoxin (PE) to make a recombinant toxin, because the new ligand-toxin junction results in improved IL4 receptor (IL4R)-binding (R. J. Kreitman et al., Proc. Natl. Acad. Sci. USA, 91: 6889- 6893, 1994). We now report that the improved binding of circularly permuted IL4-toxin is associated with improved antitumor activity in tumor-bearing mice. For in vivo testing, we made an improved circularly permuted IL4- toxin, termed IL4(38-37)-PE38KDEL. It contains an N38D mutation at the amino terminus, allowing improved expression and large-scale production in Escherichia coli. It also contains the truncated

toxin PE38KDEL, which is composed of amino acids 253364 and 381-608 of PE,
followed by ***KDEL*** . To evaluate antitumor activity, nude mice carrying s.c. tumors composed of IL4R- bearing human A431 epidermoid carcinoma cells were injected with recombinant toxins i.v. every other day for three doses. IL4(38-37)-PE38KDEL induced complete remissions in 80% of mice receiving 50 .mu.g/kg x 3 and 100% of mice receiving 100 .mu.g/kg x 3, while only 70% of mice receiving 200 .mu.g/kg x 3 of the native IL4-toxin IL4-PE38KDEL obtained complete remission. Disease-free survival after obtaining complete remissions was higher in mice treated with IL4(38-37)-PE38KDEL 50 .mu.g/Kg QOD x 3 than with IL4-PE38KDEL 200 .mu.g/Kg QOD x 3 ($P < 0.03$). IL4(38-37)-PE38KDEL and IL4-PE38KDEL exhibited similar toxicity and pharmacokinetics in the mice, indicating that the improved antitumor activity of the circularly permuted IL4-toxin was due to its improved binding to the IL4R on the target cells.

L41 ANSWER 56 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 38
AN 1996:36040 BIOSIS
DN PREV199698608175
TI Beta-2-Microglobulin with an endoplasmic reticulum retention signal increases the surface expression of folded class I major histocompatibility complex molecules.
AU Solheim, Joyce C.; Johnson, Nancy A. (1); Carreno, Beatriz M.; Lie, Wen-Rong; Hansen, Ted. H.
CS (1) Washington Univ. Sch. Med., Dep. Genetics 4566 Scott Ave., Box 8232, St. Louis, MO 63110 USA
SO European Journal of Immunology, (1995) Vol. 25, No. 11, pp. 3011-3016. ISSN: 0014-2980.
DT Article
LA English
AB With beta-2-microglobulin- (beta-2m-) cell lines such as R1E/D-b, the surface expression of class I major histocompatibility complex molecules is greatly impaired, and class I molecules that are on the Surface are generally misfolded. To determine whether beta-2m must be continually present with the class I heavy chain for the class I molecule to reach the surface in a folded conformation, a sequence encoding an endoplasmic reticulum (ER) retention signal (***KDEL***) was attached onto the 3' end of a beta-2m cDNA. After this ***chimeric*** cDNA was transfected into RIE/D-b cells, beta-2m- ***KDEL*** protein was detectable by an anti-beta-2m serum within the cells but not at the cell surface. Interestingly, RIE/D-b cells transfected with beta-2m- ***KDEL*** were found to express a high level of conformationally correct D-b molecules at the cell surface. This observation implies that beta-2m has a critical and temporal rote in the de novo folding of the class I heavy chain. We propose that the critical time for beta-2m association is when the class I molecule is docked with the transporter associated with antigen processing (TAP) and first interacts with peptide.

L41 ANSWER 57 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 39
AN 1996:24261 BIOSIS
DN PREV199698596396
TI Characterization of essential domains for the functionally of the MHBs-t transcriptional activator and identification of a minimal MHBs-t activator.
AU Hildt, Eberhard (1); Urban, Stephan; Hofschneider, Peter Hans
CS (1) Dep. Virus Res., Max-Planck-Inst. fuer Biochemie, Am Klopferspitz 18a, D-82152 Martinsried Germany
SO Oncogene, (1995) Vol. 11, No. 10, pp. 2055-2066. ISSN: 0950-9232.
DT Article
LA English
AB Integrated hepatitis B virus DNA derived from hepatocellular carcinomas can express, in one third of the cases investigated so far, a transcriptional activator encoded from 3' terminal truncated surface (preS/S) genes resulting in a C-terminally truncated middle surface protein (MHBs-t). Since MHBs-t, in contrast to the secreted MHBs, is retained in the secretory pathway at the ER, the question as to whether the retention generates the transcriptional activator function was investigated. Through ***fusion*** of MHBs to the ERretention signal ***KDEL*** , it was shown that the intracellular retention does not generate the transcriptional activator function. Tryptic digestions of microsomal vesicles revealed that the amino terminal domain of MHBs-t directs into the cytoplasmic compartment, whereas in MHBs this domain directs into the lumen of the ER. This structural difference appears to be why transcriptional activator function arises. Through deletion analysis it was shown that non-membrane-associated NHBs-t proteins are also functional activators. Nonmembrane associated MHBs-t proteins represent a second class of MHBs-t proteins. These MHBs-t-proteins are homogenously distributed all over the cell and show no difference in functionality as compared to the membrane-associated MHBs-t proteins. MHBs-t53 (truncated at aa53) was shown to be a minimal activator of this class. Both classes of MHBs-t proteins were found to form dimers; an amphiphatic alpha helix was identified within aa 41-52, which is involved in mediating the dimerization. The integrity of this domain was also revealed to be a prerequisite for the functionality of the activator, suggesting a linkage between dimerization and functionality.

L41 ANSWER 58 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 40
AN 1996:57272 BIOSIS
DN PREV199698629407
TI Anterograde and retrograde traffic between the rough endoplasmic reticulum and the Golgi complex.
AU Stinchcombe, Jane C.; Nomoto, Hiroshi; Cutler, Daniel F.; Hopkins, Colin R. (1)
CS (1) MRC Lab. Molecular Cell Biol., Dep. Biol. Biochem., Univ Coll. London, Gower St., London WC1 6BT UK
SO Journal of Cell Biology, (1995) Vol. 131, No. 6 PART 1, pp. 1387-1401. ISSN: 0021-9525.
DT Article
LA English
AB The transfer of newly synthesized membrane proteins moving from the rough endoplasmic reticulum (RER) to the Golgi complex has been studied by electron microscopy in HEp-2 cells transfected with cDNAs for ***chimeric*** proteins. These proteins consist of a reporter enzyme, horseradish peroxidase (HRP), anchored to the transmembrane domains of two integral membrane proteins, the transferrin receptor and sialyltransferase. The chimeras are distributed throughout the nuclear envelope, RER, vesicular tubular clusters (VTCs) and a network of tubules in the cis-Golgi area. At 20 degree C tubules containing chimera connect the RER to the VTCs and to the cis-Golgi network. On transfer to 37 degree C in the presence of dithiothreitol (DTT), the chimeras are seen to move from the RER and through the Golgi stack. With this temperature shift the direct connections with the RER are lost and free vesicles form; some of these vesicles contain HRP reaction product which is much more concentrated than in the adjacent RER while others lack reaction product entirely. In cells expressing SSHRP- ***KDEL*** , DAB reaction product remains distributed throughout the RER, the VTCs, and the cis-Golgi network for prolonged periods in the presence of DTT and almost all of the vesicles which form at 37 degree C are DAB-positive. Together these observations demonstrate that all three chimeras are transported from the RER to the cis-Golgi in free, 40-60-nm vesicles at 37 degree C. They also suggest that the retrograde traffic which carries SSHRP- ***KDEL*** back to the RER is probably mediated by vesicles with a similar morphology but which, in cells expressing membrane-anchored chimeras, lack detectable reaction product.

L41 ANSWER 59 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 41
AN 1995:484116 BIOSIS
DN PREV199598498416
TI Production of rat protein disulfide isomerase in Saccharomyces cerevisiae.
AU Laboissiere, Martha C. A.; Chivers, Peter T.; Raines, Ronald T. (1)
CS (1) Dep. Biochem., Univ. Wisconsin-Madison, 420 Henry Mall, Madison, WI 53706-1569 USA
SO Protein Expression and Purification, (1995) Vol. 6, No. 5, pp. 700-706. ISSN: 1046-5928.
DT Article
LA English
AB Protein disulfide isomerase (PDI) is an abundant protein of the endoplasmic reticulum that catalyzes the oxidation of protein sulfhydryl groups and the isomerization and reduction of protein disulfide bonds. Saccharomyces cerevisiae cells lacking PDI are inviable. PDI is a component of many different protein processing complexes, and the actual activity of PDI that is required for cell viability is unclear. A cDNA that codes for rat PDI fused to the alpha-factor pre-pro segment was expressed in a protease-deficient strain of S. cerevisiae under the control of an ADH2-GAPDH ***hybrid*** promoter. The cells processed the resulting protein and secreted it into the medium as a monomer, despite having a ***KDEL*** or HDEL sequence at its C-terminus. The typical yield of isolated protein was 2 mg per liter of culture. The catalytic activity of the PDI from S. cerevisiae was indistinguishable from that of PDI isolated from bovine liver. This expression system is unique in allowing the same plasmid to be used both to complement pdi1-DELTA S. cerevisiae and to produce PDI for detailed in vitro analyses. Correlations of the in vivo behavior and in vitro properties of PDI are likely to reveal structure-function relationships of biological importance.

L41 ANSWER 60 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 42
AN 1996:67012 BIOSIS
DN PREV199698639147
TI Generation of a potent ***chimeric*** toxin by replacement of domain III of Pseudomonas exotoxin with ricin A chain ***KDEL*** .
AU Pitcher, Carol; Roberts, Lynne; Fawell, Stephen; Zdanovsky, Alex G.; Fitzgerald, David J. (1); Lord, J. Michael
CS (1) Lab. Molecular Biol., DCBDC, Natl. Cancer Inst., Natl. Inst. Health, Build. 37, Room 4B03, 37 Convent Drive, MSC 24255, Bethesda, MD 20892-4255
USA
SO Bioconjugate Chemistry, (1995) Vol. 6, No. 5, pp. 624-629. ISSN: 1043-1802.
DT Article
LA English
AB Following cellular uptake, Pseudomonas exotoxin (PE) is cleaved by cellular protease which generates an enzymatically active C-terminal fragment (amino acids 280-613). This 37 kD fragment translocates to the cell cytosol where it ADP-ribosylates elongation factor 2 and inhibits protein synthesis. A recombinant ***hybrid*** toxin (designated PE-RTA) in which the ADP-ribosylation domain (domain III) was replaced by the RNA N-glycosidase domain of ricin (the A chain or RTA) has been produced in E. coli. The ***hybrid*** toxin effectively and specifically depurinated 28S ribosomal RNA, indicating that the ricin A moiety folded into its native conformation. The cytotoxicity of PE-RTA for L929 cells was approximately 100-fold less than either native PE or whole ricin. However, the addition of the tetrapeptide ***KDEL*** to the C-terminus of PE-RTA (producing PE-RTA ***KDEL***) increased cytotoxicity to the level of the native toxins. By analogy to PE, both

PE-RTA and PE-RTA ***KDEL*** would be proteolytically cleaved within PE domain H during cell entry. A single amino acid substitution, believed to disrupt an essential step in the transport of the catalytically active PE fragment to the cell cytosol (Trp281 to Ala: Zdanovsky, A.G., Chiron, M., Pastan, I., and FitzGerald, D. J. (1993) J. Biol. Chem. 268, 21791-21799), reduced the cytotoxicities of both PE and PE -RTA ***KDEL*** by approximately 100-fold. Taken together, these data show that the ricin A chain component of the ***hybrid*** toxin requires essential PE-derived sequences at both the N- and C-termini of the translocating fragment. Clearly, in the context of this ***fusion*** protein, ricin A chain cannot effect its own transfer to the cytosol.

L41 ANSWER 61 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 43
AN 1995:295591 BIOSIS
DN PREV199598309891
TI CBP-140, a novel endoplasmic reticulum resident Ca-2+-binding protein with a carboxy-terminal NDEL sequence showed partial homology with 70-kDa heat shock protein (hsp70.
AU Naved, Apala Farhat (1); Ozawa, Masayuki (1); Yu, Su (1); Miyauchi, Teruo; Muramatsu, Hisako (1); Muramatsu, Takashi (1)
CS (1) Dep. Biochemistry, Faculty Med., Kagoshima Univ., 8-35-1 Sakuragaoka, Kagoshima 890 Japan
SO Cell Structure and Function, (1995) Vol. 20, No. 2, pp. 133-141.
ISSN: 0386-7196.
DT Article
LA English
AB Antibodies against pokeweed agglutinin binding proteins isolated from F9 embryonal carcinoma cells were used to screen a lambda-gt11 expression library constructed from the cells. A cDNA clone thus obtained encoded a novel calcium binding protein of 140 kDa (CBP-140). Antibodies raised against the CBP-140 ***fusion*** protein stained a 140 kDa band in extracts not only from F9 cells but also from various mouse organs. A calcium blot experiment using CBP-140 ***fusion*** protein verified the calcium binding property of the protein. In the partial amino acid sequence so far clarified (652 amino acid residues) we could not detect EF-hand, but could detect contiguous acidic amino acids, which may serve as a calcium-binding site. CBP-140 showed homology with 70-kDa beat shock protein, though it was not induced by beat shock treatment. Localization of CBP-140 in endoplasmic reticulum was shown by indirect immunofluorescence staining and also by subcellular fractionation. Amino acid sequence of CBP-140 contains a carboxyl-terminal Asn-Asp-Glu-Leu (NDEL) sequence, which resembles Lys-Asp-Glu-Leu (***KDEL***) sequence, a signal to retain the resident proteins in endoplasmic reticulum; NDEL sequence may indeed play a similar role.

L41 ANSWER 62 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 44
AN 1995:111963 BIOSIS
DN PREV199598126263
TI Cytotoxic activity of ***chimeric*** toxins containing the epidermal growth factor-like domain of heregulins fused to PE38KDEL, a truncated recombinant form of Pseudomonas exotoxin.
AU Kihara, Ako; Pastan, Ira (1)
CS (1) Lab. Molecular Biol., Div. Cancer Biol., Diagnosis Centers, National Cancer Inst., NIH, 9000 Rockville Pike, 37/4E16, Bethesda, MD USA
SO Cancer Research, (1995) Vol. 55, No. 1, pp. 71-77.
ISSN: 0008-5472.
DT Article
LA English
AB The EGF-like domains of heregulin alpha, beta-1, beta-2, and beta-3 were fused to a truncated form of Pseudomonas exotoxin (PE38KDEL), which contains a modified carboxyl-terminal sequence, ***KDEL*** , that increases that toxin activity. The resulting ***chimeric*** toxins were produced in Escherichia coli purified to near homogeneity, and shown to be cytotoxic to target cells with very high activity on HTB20, N-87 MCF-7, and HepG2 cells; high activity on A431 and MDA-MB468 cells; and low activity toward SK-OV3, L929, and KB cells. The fact that cytotoxicity did not correlate with the levels of erbB2 expression indicated that another receptor in the erb family might be involved. Accordingly, cytotoxicity assays were performed on NIH/3T3 cell lines transfected with EGFR, ErbB2, ErbB3, or ErbB4. The results indicate that the heregulin toxins target ErbB4 or possibly ErbB3 but not ErbB2.

L41 ANSWER 63 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 45
AN 1995:222565 BIOSIS
DN PREV199598236865
TI Importance of the glutamate residue of ***KDEL*** in increasing the cytotoxicity of Pseudomonas exotoxin derivatives and for increased binding to the ***KDEL*** receptor.
AU Kreitman, Robert J.; Pastan, Ira (1)
CS (1) Lab. Mol. Biol., Natl. Cancer Inst. Health, 9000 Rockville Pike, Bethesda, MD 20892 USA
SO Biochemical Journal, (1995) Vol. 307, No. 1, pp. 29-37.
ISSN: 0264-6021.
DT Article
LA English
AB It was previously shown that amino acids 609-613 (REDLK) at the C-terminus of Pseudomonas exotoxin (PE) are necessary for cytotoxicity, presumably by directing the toxin to the endoplasmic reticulum (ER) (Chaudhary, Jinno, FitzGerald and Pastan (1990) Proc. Natl. Acad. Sci. U.S.A. 87, 308-312). Using the anti-(interleukin 2 receptor (IL2R)) immunotoxin anti-Tac(Fv)-PE38 (AT-PE38REDLK), it was found that removing the terminal lysine did not alter the activity, but replacing REDL with ***KDEL*** , the most common ER retention sequence, increased activity. To determine which amino acid in ***KDEL*** was responsible for the increase in activity, we tested eight C-terminal mutants of AT-PE38REDLK. Using IL2R-bearing MT-1 cells, we found that the glutamate residue of ***KDEL*** was required for high activity, as the cytotoxicity of AT-PE38 ending in ***KDEL*** , RDEL, KEEL or REEL was much greater than that of AT-PE38 ending in REDL, KEDL, RDDL or KDDL. Using freshly isolated lymphocytic leukaemia cells, AT-PE38 ending in ***KDEL*** . REEL or RDEL was more than 100-fold more cytotoxic than AT-PE38 ending in KEDL, REDL, RDDL or the native sequence REDLK. The RDEL sequence also improved the cytotoxic activity of an interleukin 4-PE38 toxin ***fusion*** protein. Improved cytotoxic activity correlated with improved binding of the C-termini to the ***KDEL*** receptor on rat Golgi membranes. These data indicate that the glutamate residue of ***KDEL*** improves the cytotoxicity of PE by increasing binding to a sorting receptor which transports the toxin from the transreticular Golgi apparatus to the ER, where it is translocated to the cytosol and inhibits protein synthesis.

L41 ANSWER 64 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 46
AN 1995:62233 BIOSIS
DN PREV199598076533
TI Subcellular localization and targeting of cathepsin E.
AU Finley, Elaine M.; Kornfeld, Stuart (1)
CS (1) Div. Hematol.-Oncol., Washington Univ. Sch. Medicine, 660 S. Euclid Ave, Box 8125, St. Louis, MO 63110 USA
SO Journal of Biological Chemistry, (1994) Vol. 269, No. 49, pp. 31259-31266.
ISSN: 0021-9258.
DT Article
LA English
AB The subcellular distribution and targeting of the nonlysosomal aspartic proteinase cathepsin E have been studied using mouse L cells and monkey Cos 1 cells that were transfected with cDNA encoding cathepsin E. The cathepsin E was retained in L cells for at least 20 h without significant degradation and its single N-linked oligosaccharide remained sensitive to endo-beta-N-acetylglucosaminidase H. When cathepsin E was overexpressed by transient transfection in Cos 1 cells, it was very slowly secreted into the media. The intracellular form of the enzyme contained a high mannose oligosaccharide which was processed to a complex type species upon secretion. In double label immunofluorescence studies, cathepsin E co-localized with cathepsin D-myc- ***KDEL*** , an endoplasmic reticulum (ER) marker. Subcellular fractionation on a Percoll density gradient showed that the cathepsin E co-migrated with membranous vesicles that were distinct from dense lysosomes. Only a trace amount of the enzyme was recovered in the soluble fraction. These findings indicate that in L cells and Cos 1 cells, the intracellular location of cathepsin E is the endoplasmic reticulum. To identify the protein sequences required for ER retention, we made ***chimeric*** proteins between cathepsin E and pepsinogen, an aspartic proteinase that is rapidly secreted by Cos 1 cells. We found that amino acids 1-48 of cathepsin E are important for its retention in the ER. Within this region, Cys-7, which is involved in covalent dimer formation, plays a significant role in the retention.

L41 ANSWER 65 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 47
AN 1994:367148 BIOSIS
DN PREV199497380148
TI Intracellular membrane traffic of human immunodeficiency virus type 1 envelope glycoproteins: Vpu liberates Golgi-targeted gp160 from CD4-dependent retention in the endoplasmic reticulum.
AU Kimura, Tominori (1); Nishikawa, Masao; Ohyama, Akio
CS (1) Dep. Microbiol., Kanasi Med. Univ., Moriguchi, Osaka 570 Japan
SO Journal of Biochemistry (Tokyo), (1994) Vol. 115, No. 5, pp. 1010-1020.
ISSN: 0021-924X.
DT Article
LA English
AB The membrane traffic of human immunodeficiency virus type 1 (HIV-1) envelope glycoproteins has been investigated in COS-1 cells transiently expressing the HIV-1 env, vpu, and rev genes. Analysis of oligosaccharide processing revealed that the majority of gp160 remained fully endo-H sensitive throughout a 21-h chase period, and hence cleavage of gp160 to gp120-gp41 took place prior to the creation of ***hybrid*** and complex oligosaccharides on gp120. Immunofluorescence microscopy demonstrated that in the absence of CD4 both gp160 and Vpu are targeted to the Golgi apparatus, that can be stained with wheat germ agglutinin or antibodies to the human ***KDEL*** receptor. In contrast, gp160 complexed with CD4 was retained in the ER and thus failed to reach the cis-Golgi compartment. Although gp160-bound CD4 has its own half life of 4 h 35 min in the endoplasmic reticulum (ER), co-expression of Vpu accelerated the turnover of CD4 by 5.5-fold and thereby enabled gp160 to be translocated out of the ER to the cis-Golgi compartment. We concluded that Vpu prevents the formation of stable CD4-gp160 complexes in the ER and thus indirectly allows gp160 to accumulate in the Golgi apparatus, where it is selectively retained to produce gp120-gp41.

L41 ANSWER 66 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 48
AN 1995:40634 BIOSIS
DN PREV199598054934
TI Analysis of Sequences Required for the Cytotoxic Action of a ***Chimeric*** Toxin Composed of Pseudomonas Exotoxin and Transforming Growth Factor alpha.
AU Kihara, Ako; Pastan, Ira (1)
CS (1) Lab. Mol. Biol., Div. Cancer Biol., Diagnosis Cent., Natl. Cancer Inst., Natl. Inst. Health, 9000 Rockville Pike, Build. 37, Room 4E16,

Bethesda, MD 20892 USA
SO Bioconjugate Chemistry, (1994) Vol. 5, No. 6, pp. 532-538.
ISSN: 1043-1802.
DT Article
LA English
AB ***Chimeric*** toxins composed of transforming growth factor alpha fused to mutant forms of Pseudomonas exotoxin bind to the EGF receptor and kill cells bearing these receptors. In early experiments, the binding domain of Pseudomonas exotoxin was deleted and replaced with TGF-alpha to make TGF-alpha-PE40. This ***chimeric*** toxin required proteolytic processing within the target cell to be converted to its active form (Siegall et al. (1989) FASEB J. 3, 2647-2652). Subsequently, recombinant toxins that do not require proteolytic processing were constructed by inserting TGF-alpha near the carboxyl terminus of domain III and deleting toxin residues up to the processing site at position 280. In addition, the carboxyl terminus of this toxin was converted from REDLK to ***KDEL*** to increase its activity. Recombinant toxins of this type, termed PE37/TGF-alpha/ ***KDEL*** , are about 100-fold more potent than TGF-alpha-PE40. To determine if other sequences can be removed from such ***chimeric*** toxins to make a smaller molecule that can penetrate tissues better, we have carried out a deletion analysis of sequences present within domains II and Ib. We find that all of domain Ib and a portion of domain II can be deleted without significant loss of cytotoxic activity, but larger deletions extending further into domain II lose cytotoxic activity. We also find that inserting a small linking peptide (Gly)-4Ser between residual sequences in domain II and domain III, in molecules with diminished cytotoxic activity, enhances cytotoxicity suggesting that one role of domain Ib is to prevent undesirable interactions between domains II and III. These new ***chimeric*** toxins are very active on A431 epidermoid carcinoma cells which contain many EGF receptors. One of these was also tested in animals and showed strong antitumor activity against A431 tumors growing in nude mice.

L41 ANSWER 67 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS
AN 1995:53247 BIOSIS
DN PREV199598067547
TI The ***KDEL*** -receptor accumulates after brefeldin A treatment in vesicular structures (BIVS) which are distinct from the ER-Golgi ***hybrid*** compartment.
AU Fuellekrug, J.; Mieskes, G.
CS Dep. Clin. Biochem., Univ. Goettingen, 37075 Goettingen Germany
SO Molecular Biology of the Cell, (1994) Vol. 5, No. SUPPL., pp. 439A. Meeting Info.: Thirty-fourth Annual Meeting of the American Society for Cell Biology San Francisco, California, USA December 10-14, 1994 ISSN: 1059-1524.
DT Conference
LA English

L41 ANSWER 68 OF 92 CAPLUS COPYRIGHT 2001 ACS
AN 1995:67090 CAPLUS
DN 122:31946
TI Synthesis of peptide-oligonucleotide hybrids containing a ***KDEL*** signal sequence
AU Arar, K.; Monsigny, M.; Mayer, R.
CS Cent. Biophys. Mol., CNRS, Orleans, F-45071, Fr.
SO Pept.: Chem., Struct. Biol., Proc. Am. Pept. Symp., 13th (1994), Meeting Date 1993, 184-6. Editor(s): Hodges, Robert S.; Smith, John A. Publisher: ESCOM, Leiden, Neth.
CODEN: 60LXAW
DT Conference
LA English
GI

/ Structure 1 in file .gra /

AB A symposium report on the synthesis of peptide-oligonucleotide hybrids contg. a ***KDEL*** signal sequence by linking a 3'-thiol oligonucleotide to a N.alpha.-maleimidocaproyl peptide. The oligonucleotide used is a 12-mer with a sequence specific for Ha-ras around the point mutation in the 12th codon. Thus, H-Tyr-Lys-Asp-Glu-Leu-OH was converted into the N.alpha.-maleimidocaproyl deriv., which was treated with 3'-thiol oligonucleotide to give peptide-oligonucleotide ***hybrid*** I.

L41 ANSWER 69 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 49
AN 1994:180820 BIOSIS
DN PREV199497193820
TI In vivo activities of acidic fibroblast growth factor-Pseudomonas exotoxin ***fusion*** proteins.
AU Siegall, Clay B. (1); Gawlak, Susan L.; Chace, Dana F.; Merwin, June R.; Pastan, Ira
CS (1) Bristol-Myers Squibb, Pharmaceutical Res. Inst., Molecular Immunology Dep., 3005 First Avenue, Seattle, WA 98121 USA
SO Bioconjugate Chemistry, (1994) Vol. 5, No. 1, pp. 77-83.
ISSN: 1043-1802.
DT Article
LA English
AB Fibroblast growth factor receptors are highly expressed in a variety of cancer cells and activated vasculature. Using ***chimeric*** toxins targeted to cell-surface aFGF receptors, we have demonstrated specific cytotoxic activity to these cell types. These molecules, aFGF-PE40 and aFGF-PE4E ***KDEL*** , are ***fusion*** proteins containing acidic FGF and either a 40- or a 66-kDa binding defective form of Pseudomonas exotoxin, respectively. Both aFGF-toxin ***fusion*** proteins were able to inhibit protein synthesis in vitro in a variety of carcinoma cell lines. The half-life of aFGF-PE40 in serum was found to be 41 min when coadministered with heparin. Administration of aFGF-PE40 or aFGF-PE4E ***KDEL*** with heparin inhibits the growth of established KB and preestablished A431 epidermoid carcinoma xenografts in athymic mice. The antitumor activities of the two aFGF-toxin ***fusion*** proteins were equivalent against the KB tumor xenografts. While we were able to slow the growth of the KB tumor xenografts, we were unable to cause tumor regressions. Histochemical analysis of treated versus untreated tumor tissue revealed a difference in tumor size but not of vascularity. We conclude that aFGF-PE40 and aFGF-PE4E ***KDEL*** have in vivo antitumor activity that targets the tumor cell mass rather than vascular structures in mice xenografted with human epidermoid carcinoma.

L41 ANSWER 70 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 50
AN 1993:520353 BIOSIS
DN PREV199396133760
TI Transmembrane topology of the mammalian ***KDEL*** receptor.
AU Singh, Paramjeet; Tang, Bor Luen; Wong, Siew Heng; Hong, Wanjin (1)
CS (1) Membrane Biol. Lab., Inst. Mol. Cell Biol., National University Singapore, Sinapgore 0511 Singapore
SO Molecular and Cellular Biology, (1993) Vol. 13, No. 10, pp. 6435-6441.
ISSN: 0270-7306.
DT Article
LA English
AB The mammalian ***KDEL*** receptor is an integral membrane protein with seven hydrophobic regions. ***Fusion*** proteins comprising a 37-kDa N-glycosylation reporter fused downstream of amino-terminal fragments of the ***KDEL*** receptor with varying numbers of hydrophobic regions were synthesized in an in vitro translation system containing canine pancreatic microsomes. The luminal or cytosolic orientation of the reporter, and hence of the hydrophilic region to which it is fused, was inferred from the presence or absence of glycosylation, which occurs only in the lumen of the microsomes. The cytosolic orientation of the N and C termini was also confirmed immunocytochemically. Our results suggest that the ***KDEL*** receptor is inserted into the membrane with only six transmembrane domains and that both the amino and carboxy termini are located in the cytoplasm.

L41 ANSWER 71 OF 92 EMBASE COPYRIGHT 2001 ELSEVIER SCI. B.V.
AN 93081604 EMBASE
DN 1993081604
TI Heparin-binding transforming growth factor .alpha.-Pseudomonas exotoxin A. A heparan sulfate-modulated recombinant toxin cytotoxic to cancer cells and proliferating smooth muscle cells.
AU Mesri E.A.; Kreitman R.J.; Fu Y.-M.; Epstein S.E.; Pastan I.
CS Lab. of Molecular Biology, National Cancer Institute, National Institutes of Health, 9000 Rockville Pike,Bethesda, MD 20892, United States
SO Journal of Biological Chemistry, (1993) 268/7 (4853-4862).
ISSN: 0021-9258 CODEN: JBCHA3
CY United States
DT Journal; Article
FS 004 Microbiology
016 Cancer
029 Clinical Biochemistry
052 Toxicology
LA English
SL English
AB TGF.alpha.-PE40, a recombinant toxin in which transforming growth factor .alpha. (TGF.alpha.) is fused to a mutant form of Pseudomonas exotoxin, is selectively cytotoxic to cells bearing epidermal growth factor (EGF) receptors. Heparin binding EGF-like growth factor is a potent mitogen for smooth muscle cells capable of binding to both the EGF receptor and to immobilized heparin (Higashiyama, S., Abraham, J., Miller, J., Fiddes, J., and Klagsbrun, M. (1991) Science 251, 936-938). To study the effect of the heparin-binding domain in a ***chimeric*** toxin targeted to the EGF receptor, we fused the DNA sequence corresponding to the putative NH2-terminal heparin-binding (HB) domain of HB-EGF to ***chimeric*** toxins composed of TGF.alpha. and two different recombinant forms of Pseudomonas exotoxin (PE). One of these is a truncated form of PE devoid of the binding domain (TGF.alpha.-PE38); another is a mutant form of full-length toxin containing inactivating mutations in the binding domain and an altered carboxyl terminus (TGF.alpha.-PE(4E) ***KDEL***). The resulting ***chimeric*** toxins HB-TGF.alpha.-PE38 and HB-TGF.alpha.-PE(4E) ***KDEL*** were expressed in Escherichia coli as inclusion bodies, refolded, and purified by heparin affinity chromatography. Both of the toxins were eluted from heparin at 0.8 M NaCl, in contrast to their respective TGF.alpha. toxins which were eluted at 0.15 M. Binding studies on A431 cells showed that the HB-TGF.alpha. toxins bound to the EGF receptor with an affinity similar to that of the TGF.alpha. toxins. However, cell killing studies on a panel of malignant cell lines showed that cytotoxicity was strongly affected by the presence of the HB domain. Cell lines expressing high numbers of EGF receptors such as A431 and KB were less sensitive to toxins containing the HB domain. Cells with low number of EGF receptors had similar responses to both types of toxins (MCF-7 and LNCaP) or were more sensitive to the toxin with the added HB domain (HEP-G2). HB- TGF.alpha.-PE(4E) ***KDEL*** was over 10-fold more cytotoxic against proliferating vascular smooth muscle cells (VSMC) than to quiescent VSMC. Moreover, HB- TGF.alpha.-PE(4E) ***KDEL***

was 6-fold more potent than TGF.alpha.-PE(4E) ***KDEL*** to proliferating VSMC. Competition studies with EGF and/or heparin showed that heparin blocks the cytotoxicity of HB-TGF toxins and the inhibitory action of heparin is stronger in cells expressing lower number of EGF receptors. In addition, experiments with heparitinase-treated cells showed that in cells with low numbers of EGF receptors the binding of the HB domain to cell surface heparan sulfate proteoglycans appears to facilitate the internalization of the toxin. We conclude that addition of a HB domain to TGF.alpha.-PE38 or TGF.alpha.-PE(4E) ***KDEL*** confers the ability to bind to and to be modulated by heparin-like molecules and increases their cytotoxicity to cells expressing low numbers of EGF receptor and proliferating smooth muscle cells.

L41 ANSWER 72 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 51
AN 1994:18532 BIOSIS
DN PREV199497031532
TI Quality control of ER synthesized proteins: An exposed thiol group as a three-way switch mediating assembly, retention and degradation.
AU Fra, Anna M.; Fagioli, Claudio; Finazzi, Dario; Sitia, Roberto (1); Alberini, Cristina M.
CS (1) DIBIT-HSR, Milano Italy
SO EMBO (European Molecular Biology Organization) Journal, (1993) Vol. 12, No. 12, pp. 4755-4761.
ISSN: 0261-4189.
DT Article
LA English
AB Plasma cells secrete IgM only in the polymeric form: the C-terminal cysteine of the mu heavy chain (Cys575) is responsible for both intracellular retention and assembly of IgM subunits. Polymerization is not quantitative, and part of IgM is degraded intracellularly. Neither chloroquine nor brefeldin A (BFA) inhibits degradation, suggesting that this process occurs in a pre-Golgi compartment. Degradation of IgM assembly intermediates requires Cys575: the monomeric IgMala575 mutant is stable also when endoplasmic reticulum (ER) to Golgi transport is blocked by BFA. Addition of the 20 C-terminal residues of mu to the lysosomal protease cathepsin D is sufficient to induce pre-Golgi retention and degradation of the ***chimeric*** protein: the small amounts of molecules which exit from the ER are mostly covalent dimers. By contrast, when retained by the ***KDEL*** sequence, cathepsin D is stable in the ER, indicating that retention is not sufficient to cause degradation. Replacing the C-terminal cysteine with serine restores transport through the Golgi. As all ***chimeric*** cathepsin D constructs display comparable protease activity in vitro, their different fates are not determined by gross alterations in folding. Thus, also out of its normal context, the mu chain Cys575 plays a crucial role in quality control, mediating assembly, retention and degradation.

L41 ANSWER 73 OF 92 EMBASE COPYRIGHT 2001 ELSEVIER SCI. B.V.DUPLICATE 52
AN 93127527 EMBASE
DN 1993127527
TI Recombinant immunotoxins containing the V(H) or V(L) domain of monoclonal antibody B3 fused to Pseudomonas exotoxin.
AU Brinkmann U.; Lee B.K.; Pastan I.
CS Laboratory of Molecular Biology, NCI, NIH, 9000 Rockville Pike,Bethesda, MD 20892, United States
SO Journal of Immunology, (1993) 150/7 (2774-2782).
ISSN: 0022-1767 CODEN: JOIMA3
CY United States
DT Journal; Article
FS 004 Microbiology
026 Immunology, Serology and Transplantation
029 Clinical Biochemistry
LA English
SL English
AB We prepared recombinant immunotoxins in Escherichia coli in which the V(H) or V(L) domains of mAb B3 were fused to a truncated form of Pseudomonas exotoxin (PE) (PE38KDEL). mAb B3 binds to a carbohydrate Ag found on the surfaces of many types of cancers and only a few normal tissues. PE38KDEL is a 38-kDa form of PE (66 kDa) that is missing the cell-binding domain of PE and has the carboxyl end changed from REDLK to ***KDEL*** . We show that immunotoxins in which the H chain or the L chain V region is fused to PE38KDEL bind to and kill carcinoma cells containing the B3 Ag. B3 Ag-negative cells were not affected. The cytotoxicity of these molecules is between 20- and 100-fold less than B3(Fv)-immunotoxins, containing both the H and L chain V regions. The V(L)-containing toxin was more active than the V(H)-containing toxin, indicating that the L chain of mAb B3 probably contributes more to Ag-binding than the H chain. Refolding experiments show that B3(V(L))-PE38KDEL aggregates less than the V(H)-derivative or than a single chain immunotoxin B3(Fv)-PE38KDEL, which contains both domains in a single chain form. Furthermore, in addition to monomers, active homodimers of B3(V(H))- and B3(V(L))-PE38KDEL were obtained from renaturation experiments. The V(L)-toxin dimers, which might have their binding regions arranged in a manner similar to Bence Jones proteins (L chain homodimers), were found to have almost the same cytotoxicity as the monomers, whereas the V(H)-toxin dimers had decreased cytotoxic activity.

L41 ANSWER 74 OF 92 EMBASE COPYRIGHT 2001 ELSEVIER SCI. B.V.
AN 93297551 EMBASE
DN 1993297551
TI Cytotoxic effects of vascular smooth muscle cells of the ***chimeric*** toxin, heparin binding TGF.alpha.-Pseudomonas exotoxin.
AU Fu Y.-M.; Mesri E.A.; Yu Z.-X.; Kreitman R.J.; Pastan I.; Epstein S.E.
CS Nat. Heart, Lung and Blood Institute, National Institutes of Health,Bethesda, MD 20892, United States
SO Cardiovascular Research, (1993) 27/9 (1691-1697).
ISSN: 0008-6363 CODEN: CVREAU
CY United Kingdom
DT Journal; Article
FS 002 Physiology
018 Cardiovascular Diseases and Cardiovascular Surgery
030 Pharmacology
037 Drug Literature Index
LA English
SL English
AB Objective: Smooth muscle cell proliferation appears to be very important in restenosis after angioplasty. A ***chimeric*** toxin created by genetically fusing the gene encoding TGF.alpha. (targets the EGF receptor) to the gene encoding Pseudomonas exotoxin (PE) preferentially kills rapidly proliferating smooth muscle cells. Recently, a heparin binding EGF-like growth factor (HB-EGF) has been identified. The HB domain enhances the mitogenic activity for smooth muscle cells. The purpose of this study was to design a new ***chimeric*** toxin, having both heparin binding and EGF receptor binding function, and to determine whether it is more cytotoxic to smooth muscle cells. Methods: By recombinant DNA techniques, a new ***chimeric*** toxin, HB-TGF.alpha.-PE(4E) ***KDEL*** , was synthesised. Cytotoxic assays were performed by assessing the capacity to inhibit protein synthesis of rat vascular smooth muscle cells. Results: The toxin preferentially killed rapidly proliferating smooth muscle cells ($p<0.025$). The HB domain increased the cytotoxicity of the molecule when compared to the other ***chimeric*** toxins tested against smooth muscle cells. The cytotoxic effect of the new molecule was significantly decreased by exogenously added heparin ($p<0.05$). Conclusions: The presence of a heparin binding domain increases the smooth muscle cell cytotoxicity of the TGF.alpha. ***fusion*** toxin, perhaps because HB-TGF.alpha.-PE(4E) ***KDEL*** functions as a molecule with two ligands. It will be important to determine whether the greater smooth muscle cell cytotoxicity that exists in vitro will facilitate the specific targeting and killing of rapidly proliferating cells in vivo.

L41 ANSWER 75 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 53
AN 1993:390893 BIOSIS
DN PREV199396066193
TI A recombinant form of Pseudomonas exotoxin A containing transforming growth factor alpha near its carboxyl terminus for the treatment of bladder cancer.
AU Theuer, Charles P.; Fitzgerald, David J.; Pastan, Ira (1)
CS (1) Lab. Molecular Biol., Natl. Cancer Inst., Div. Cancer Biol., Diagnosis Centers, Natl. Inst. Health, Build 37, Room 4E16, Bethesda, MD 20892
SO Journal of Urology, (1993) Vol. 149, No. 6, pp. 1626-1632.
ISSN: 0022-5347.
DT Article
LA English
AB The epidermal growth factor receptor (EGFR) is overexpressed on the superficial layers of malignant urothelium and is suspected of playing a role in tumour progression. TP40 is a ***chimeric*** protein composed of transforming growth factor-alpha (TGF-alpha) fused to a modified form of Pseudomonas exotoxin A (PE) that is selectively cytotoxic to EGFR-bearing cells and is currently undergoing clinical study for the intravesical therapy of bladder cancer. We constructed a recombinant toxin PE35/TGF-alpha- ***KDEL*** as an improved agent for the local therapy of EGFR-bearing bladder cancer. PE35/TGF-alpha- ***KDEL*** does not require intracellular proteolysis to generate a carboxyl-terminal fragment capable of reaching the target cell cytosol and contains a modified carboxyl-terminal sequence ***KDEL*** , that increases toxin activity. These features make PE35/TGF-alpha- ***KDEL*** from 10-to 700-fold more potent than TP40 on four human bladder cancer cell lines. PE35/TGF-alpha- ***KDEL*** may be a useful agent for treatment of EGFR-bearing cancers.

L41 ANSWER 76 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 54
AN 1994:64895 BIOSIS
DN PREV199497077895
TI Basic fibroblast growth factor-Pseudomonas exotoxin ***chimeric*** proteins; Comparison with acidic fibroblast growth factor-Pseudomonas exotoxin.
AU Gawlak, Susan L.; Pastan, Ira; Siegall, Clay B. (1)
CS (1) Bristol-Myers Squibb, Pharmacueitcal Res. Inst., Molecular Immunology Dep., 3005 First Avenue, Seattle, WA 98121 USA
SO Bioconjugate Chemistry, (1993) Vol. 4, No. 6, pp. 483-489.
ISSN: 1043-1802.
DT Article
LA English
AB We have constructed growth factor-toxin ***chimeric*** molecules composed of basic fibroblast growth factor (bFGF) and two different binding mutant forms of Pseudomonas exotoxin termed bFGF-PE40 and bFGF-PE4E ***KDEL*** . The ***chimeric*** molecules were expressed in Escherichia coli and localized to both inclusion bodies and the spheroplast cytoplasm. The bFGF-toxin ***fusion*** protein that was isolated and purified from inclusion bodies was 3-fold more active in inhibiting protein synthesis than that purified from spheroplast cytoplasm. Immunoreactivity of purified bFGF-toxin ***fusion*** protein to anti-bFGF antibodies was similar to that of native bFGF, as determined by ELISA analysis. A variety of carcinoma cell lines were sensitive to bFGF-PE40 and bFGF-PE4E ***KDEL*** , including H3396

(breast), Hep G2 (hepatocellular), and A431 (epidermoid). The concentration of ***chimeric*** toxin that inhibited protein synthesis by 50% (EC-50) was 110, 70, and 18 ng/mL for bFGF-PE40 and 15, 1, and 18 ng/mL for bFGF-PE4E ***KDEL*** . In comparison with ***fusion*** -toxins composed of acidic fibroblast growth factor (aFGF) and either PE40 or PE4E ***KDEL*** , bFGF-PE40 and bFGF-PE4E ***KDEL*** were similarly cytotoxic on most cell lines tested. Human aortic smooth muscle cells were sensitive to both bFGF and aFGF toxin ***fusion*** proteins. However, human aortic endothelial cells were sensitive to the bFGF-toxins but were resistant to both aFGF-toxin forms. Time course studies showed that bFGF-PE40 needed a 4-6-h exposure to target cells for peak inhibition of protein synthesis on both MCF-7 and A431 cells, while aFGF-PE40 was almost fully active within a 2-h incubation. The addition of heparin competed for the cytotoxic activity of bFGF-PE40 but not for aFGF-PE40 on MCF-7 and A431 cells. Cytotoxic forms of bFGF and aFGF may be useful in eliminating populations of cells that express both or one of these FGF receptors.

L41 ANSWER 77 OF 92 CAPLUS COPYRIGHT 2001 ACS
AN 1993:139227 CAPLUS
DN 118:139227
TI Single-chain immunotoxin fusions between anti-tac and Pseudomonas exotoxin: Relative importance of the two toxin disulfide bonds
AU Kreitman, Robert J.; Batra, Janendra K.; Seetharam, Saraswathy; Chaudhary, Vijay K.; FitzGerald, David J.; Pastan, Ira
CS Div. Cancer Biol., Natl. Cancer Inst., Bethesda, MD, 20892, USA
SO Bioconjugate Chem. (1993), 4(2), 112-20
CODEN: BCCHES; ISSN: 1043-1802
DT Journal
LA English
AB Anti-Tac(Fv)-PE40 is a recombinant single-chain immunotoxin in which the variable heavy and light domains of the anti-IL2 receptor antibody, anti-Tac, are connected to each other by a peptide linker and then fused to PE40, a truncated form of Pseudomonas exotoxin (PE). This ***fusion*** protein has four disulfide bonds: one in each of the two variables domains, one in domain II (Cys 265-287), and one in domain Ib (Cys 372-379) of PE. To study the importance of the disulfide bonds of the toxin to the activity of single-chain immunotoxins, we constructed mutants in which either the cysteines in the toxin were changed to alanines or the amino acids 365-380 of PE were deleted. We began this study with anti-Tac(Fc)-PE40 and a more active variant, anti-Tac(Fv)-PE40KDEL, in which the carbonyl terminus is changed from REDLK to ***KDEL*** . From these proteins we made anti-Tac(Fv)-PE404A and anti-Tac(Fv)-PE40KDEL4A, resp., by converting cysteines at amino acids 265, 287, 372, and 379 of PE to alanines. This change resulted in a 20-100-fold loss of activity toward human target cells, but no significant change in binding affinity to p55. To det. the importance of the second toxin disulfide bond, we removed amino acids 365-380 from anti-Tac(Fv)-PE40, anti-Tac(Fv)-PE40KDEL, and anti-Tac(Fv)-PE40KDEL4A, resulting in anti-Tac(Fv)-PE38, anti-Tac(Fv)-PE38KDEL, and anti-Tac(Fv)-PE38KDEL2A, resp. This deletion resulted in a slight increase in cytotoxicity toward some target cells. Anti-Tac(Fv)-PE38KDEL and anti-Tac(Fv)-PE40KDEL were up to 300-fold more cytotoxic than their resp. mutants which contained alanines only at positions 265 and 287. Thus the first disulfide bond of the toxin (Cys 265-287) is much more important for cytotoxicity than the second one (Cys 372-379). We found that anti-Tac(Fv)-PE40KDEL and anti-Tac(Fv)-PE38KDEL were the most active agents in vitro and had the same half-life in mice and the max. tolerated dose was also the same for each.

L41 ANSWER 78 OF 92 CAPLUS COPYRIGHT 2001 ACS
AN 1993:161049 CAPLUS
DN 118:161049
TI Recombinant immunotoxins containing monoclonal antibody B3 Fv region fused with Pseudomonas exotoxin PE40 for treating cancer
IN Pastan, Ira; Willingham, Mark; Fitzgerald, David; Brinkmann, Uli; Pai, Lee
PA United States Dept. of Health and Human Services, USA
SO U. S. Pat. Appl., 28 pp. Avail. NTIS Order No. PAT-APPL-7-767,331.
CODEN: XAXXAV
DT Patent
LA English
FAN.CNT 7

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI US 767331	A0	19921215	US 1991-767331	19910930
WO 9307286	A1	19930415	WO 1992-US8257	19920929
W: AU, CA, JP				
RW: AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LU, MC, NL, SE				
AU 9227798	A1	19930503	AU 1992-27798	19920929
AU 675413	B2	19970206		
EP 610288	A1	19940817	EP 1992-921866	19920929
EP 610288	B1	20000315		
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LI, LU, MC, NL, SE				
JP 07502643	T2	19950323	JP 1992-506994	19920929
AT 190659	E	20000415	AT 1992-921866	19920929
US 5608039	A	19970304	US 1994-331398	19941028
US 5889157	A	19990330	US 1994-331396	19941028
US 5981726	A	19991109	US 1994-331397	19941028
US 5990296	A	19991123	US 1996-759804	19961203
US 6287562	B1	20010911	US 1999-227693	19990108
PRAI US 1990-596289	A2	19901012		
US 1991-767331	A	19910930		
WO 1992-US8257	A	19920929		
US 1994-331396	A3	19941028		
US 1994-331398	A3	19941028		

AB Recombinant immunotoxins for treating cancer comprise the Fv region of monoclonal antibody B3 (to mucinous carcinomas) fused with Pseudomonas exotoxin PE40 or deriv. PE38 having C-terminus ***KDEL*** , B3(Fv)-PE40 and B3(Fv)-PE38KDEL, resp. Recombinant prepn. of B3(Fv)-PE40 and B3(Fv)-PE38KDEL is described. Injection of 2.5-10 .mu.g B3(Fv)-PE38KDEL twice daily into nude mice bearing human epidermoid carcinomas (from A431 cells) produced complete tumor regression.

L41 ANSWER 79 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 55
AN 1992:500348 BIOSIS
DN BA94:118873
TI INTERACTION OF SECRETED INSULIN-LIKE GROWTH FACTOR-I IGF-I WITH CELL SURFACE RECEPTORS IS THE DOMINANT MECHANISM OF IGF-I'S AUTOCRINE ACTIONS.
AU DAI Z; STILES A D; MOATS-STAATS B; VAN WYK J J; D'ERCOLE A J
CS DEP. PEDIATRICS, CB 7220, UNIVERSITY NORTH CAORLINA, CHAPEL HILL, NC 27599-7220.
SO J BIOL CHEM, (1992) 267 (27), 19565-19571.
CODEN: JBCHA3. ISSN: 0021-9258.
FS BA; OLD
LA English
AB In a prior report we represented evidence that insulin-like growth factor-I (IGF-I) can act in autocrine fashion by demonstrating that FRTL-5 cells transfected with hIGF-IA ***fusion*** genes express and secrete biologically active IGf-I that renders the stimulation of DNA syntehsis in FRTL-5 cells independent of their requirement for exogenous IIGFs or insulin. To determine if IGF-I's autocrine actions require secretion or can be mediated by interactions with intracellular receptors, we have created a new line of FRTL-5 cells that express a mutant IGF-IA precursor containing the endoplasmic reticulum retention amino acid sequence, Lys-Asp-Glu-Leu (***KDEL***), at its carboxyl terminus. The mutant IGF-IA/ ***KDEL*** precusor expressed by stably transfected FRTL-5 cells was shown to be retained intracellularly and to have biological activity comparable with mature IGF-I, as judged by the activity of partially purified IGF-IA/ ***KDEL*** in wild type FRTL-5 cells. Expression of IGF-IA/ ***KDEL*** in FRTL-5 cells, however, neither augmented TSH-stimulated DNA synthesis nor stimulated IGF-binding protein-5 exprssion, as does IGF-IA expression in transfected FRTL-5 cells and the addition of exogenous IGF-I to wild type FRTL-5 cells. IGF-IA/ ***KDEL*** expression, however, desensitized FRTL-5 cells to the actions of exogenous IGF-I despite having only minimal effects on cell surface type I receptor number, suggesting that intracellular IGF-I is capable of significant biological actions. The failure of IGF-IA/ ***KDEL*** to replicate the actions of secreted IGF-I, taken together with the findings that a monoclonal antibody against IGF-I blocked IGF-I's actions in IGF-I-secreting transfected FRTL-5 cells, provides evidence that IGF-I secretion and interaction with cell surface type I IGF receptors is the dominant mechanism of IGF-I's autocrine actions.

L41 ANSWER 80 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 56
AN 1992:324542 BIOSIS
DN BA94:26383
TI DIFFERENT SORTING OF LYS-ASP-GLU-LEU PROTEINS IN RAT LIVER.
AU PETER F; NGUYEN VAN P; SOLING H-D
CS ABTEILUNG KLINISCHE BIOCHEMIE, ZENTRUM INNERE MEDIZIN, UNIVERSITAET GOETTINGEN, ROBERT-KOCH-STRASSE 40, D-3400 GOETTINGEN, GER.
SO J BIOL CHEM, (1992) 267 (15), 10631-10637.
CODEN: JBCHA3. ISSN: 0021-9258.
FS BA; OLD
LA English
AB Most of the resident soluble proteins of the endoplasmic reticulum (ER) seem to be sorted into this compartment via their COOH-terminal tetrapeptide Lys-Asp-Glu-Leu (***KDEL***). This sorting is supposed to occur in a post-ER compartment. Three resident soluble ER glycoproteins belonging to the ***KDEL*** family are CaB1, CaBP2, CABP3 (= calreticulin), and CaBP4 (= grp94) (Nguyen Van, P., Peter, F., and Soling, H.-D. (1989) J. Biol. Chem. 264, 17494-17501). In rat liver, calreticulin possesses a carbohydrate moiety of the complex ***hybrid*** type with terminal galactoses (Nguyen Van, P., Peter, F., and Soling, H.-D. (1989) J. Biol. Chem. 264, 17494-17501). We can show now that practically all calreticulin molecules (and not only a fraction) possess terminal galactoses as well as the COOH-terminal ***KDEL*** sequence. This as well as pulse-chase experiments performed at 37 and 15.degree. C indicate that calreticulum must have passed through the trans Golgi. Subcellular fractionations of post-mitochondrial supernatants from isolated rat hepatocytes by sucrose-Nycodenz gradient centrifugation revealed that calreticulin is confined mainly to the rough ER, grp94 mainly to the smooth ER. CaBP1, a member of the thioredoxin family, was recovered in fractions which most likely represent the intermediate compartment. This indicates that ***KDEL*** is a sorting signal which leads to the retention of these proteins in the pre-Golgi compartments. However, additional factors, most likely residing within the specific ***KDEL*** protein itself, determine the final location of the protein within the pre-Golgi compartments. This is underlined by experiments in which the density dependent distribution of total ***KDEL*** proteins was studied using a COOH-terminal ***KDEL*** -specific antibody.

L41 ANSWER 81 OF 92 EMBASE COPYRIGHT 2001 ELSEVIER SCI. B.V.
AN 92292182 EMBASE
DN 1992292182
TI Acidic fibroblast growth factor-Pseudomonas exotoxin ***chimeric*** protein elicits antiangiogenic effects on endothelial cells.
AU Merwin J.R.; Lynch M.J.; Madri J.A.; Pastan I.; Siegall C.B.
CS TargeTech Inc., 290 Pratt Street,Meriden, CT 06450, United States
SO Cancer Research, (1992) 52/18 (4995-5001).
ISSN: 0008-5472 CODEN: CNREA8
CY United States
DT Journal; Article
FS 004 Microbiology
026 Immunology, Serology and Transplantation
029 Clinical Biochemistry
LA English
SL English
AB It has recently been shown that ***chimeric*** toxins composed of acidic fibroblast growth factor fused to mutant forms of Pseudomonas exotoxin (aFGF- PE) are cytotoxic to a variety of tumor cell lines with FGF receptors. Although aFGF-PE might be considered as a possible chemotherapeutic toxin, limited knowledge is available concerning its effect on endothelia. This study investigates whether one of the aFGF-PE ***fusion*** proteins, aFGF- PE66(4Glu) ***KDEL*** , can function as an anti-angiogenic agent. Protein synthesis studies using rat epididymal fat pad microvascular endothelial cells (RFCs) indicated that after 24 h in culture, aFGF-PE had a significant inhibitory effect on protein synthesis at concentrations >100 ng/ml. In cultures incubated with 1000 ng/ml aFGF-PE, RFC protein synthesis was inhibited as much as 83%. RFCs were also cultured in a 3-dimensional type I collagen gel and incubated with either transforming growth factor .beta.1, aFGF-PE, or a combination of both. Transforming growth factor .beta.1 elicits in vitro angiogenesis in these 3-dimensional cultures which consist of rapid formation of complex tubular structures. Transforming growth factor .beta.1-treated RFCs incubated with aFGF-PE were unable to produce this angiogenic response, nor were they able to migrate out of the 3-dimensional culture to form a monolayer as shown by controls. Cell viability analyses showed that aFGF-PE produced a dose-dependent toxic effect which ranged from 10 to 90% cell death. Competition assays in which the ***chimeric*** toxin was preincubated with antibodies to aFGF resulted in an 89% reversal of the inhibitory effects of aFGF-PE on endothelial cells. Acidic FGF-PE with a mutation in the ADP ribosylation domain of PE was inactive in both 2-dimensional and 3- dimensional cultures. These data show that aFGF-PE has specific in vitro cytotoxic, antiangiogenic, and antimigratory effects on microvascular endothelia.

L41 ANSWER 82 OF 92 EMBASE COPYRIGHT 2001 ELSEVIER SCI. B.V.
AN 92170487 EMBASE
DN 1992170487
TI Plant and mammalian sorting signals for protein retention in the endoplasmic reticulum contain a conserved epitope.
AU Denecke J.; De Rycke R.; Botterman J.
CS University of Agricultural Sciences, Uppsala Genetic Centre, Department of Molecular Genetics, Box 7003,S-75007 Uppsala, Sweden
SO EMBO Journal, (1992) 11/6 (2345-2355).
ISSN: 0261-4189 CODEN: EMJODG
CY United Kingdom
DT Journal; Article
FS 029 Clinical Biochemistry
LA English
SL English
AB We studied protein sorting signals which are responsible for the retention of reticuloplasmins in the lumen of the plant endoplasmic reticulum (ER). A non-specific passenger protein, previously shown to be secreted by default, was used as a carrier for such signals. Tagging with C-terminal tetrapeptide sequences of mammalian (***KDEL***) and yeast (HDEL) reticuloplasmins led to effective accumulation of the protein chimeras in the lumen of the plant ER. Some single amino acid substitutions within the tetrapeptide tag (SDEL, -KDDL, -KDEI and -KDEV) can cause a complete loss of its function as a retention signal, demonstrating the high specificity of the retention machinery. However, other modifications confer efficient (-RDEL) or partial (-KEEL) retention. It is also shown that the efficiency of protein retention is not significantly impaired by an increased ligand concentration in plants. The efficiently retained chimeras (- ***KDEL*** , -HDEL and -RDEL) were shown to be recognized by a monoclonal antibody directed against the C-terminus of the mammalian reticuloplasmin protein disulfide isomerase (PDI). The recognized epitope is also present in several putative reticuloplasmins in microsomal fractions of plant and mammalian cells, suggesting that the antibodies recognize an important structural determinant of the retention signal. In addition, data are discussed which support the view that upstream sequences beyond the C-terminal tetrapeptide can influence or may be part of the structure of reticuloplasmin retention signals.

L41 ANSWER 83 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 57
AN 1992:280227 BIOSIS
DN BA94:4877
TI ANALYSIS OF THE BIP GENE AND IDENTIFICATION OF AN ER RETENTION SIGNAL IN SCHIZOSACCHAROMYCES-POMBE.
AU PIDOUX A L; ARMSTRONG J
CS MEMBRANE MOL. BIOL. LAB., IMPERIAL CANCER RES. FUND, BOX 123, LINCOLN'S INN FIELDS, LONDON, WC2A 3PX, UK.
SO EMBO (EUR MOL BIOL ORGAN) J, (1992) 11 (4), 1583-1591.
CODEN: EMJODG. ISSN: 0261-4189.
FS BA; OLD
LA English
AB We have cloned the gene for the resident luminal ER protein BiP from the fission yeast, Schizosaccharomyces pombe. The predicted protein product is equally divergent from the budding yeast and mammalian homologues. Disruption of the BiP gene in S. pombe is lethal and BiP mRNA levels are regulated by a variety of stresses including heat shock. Immunofluorescence of cells expressing an epitope-tagged BiP protein show it to be localized to the nuclear envelope, around the cell periphery and in a reticular structure through the cytoplasm. Unexpectedly, we find the BiP protein contains an N-linked glycosylation site which can be utilized. The C-terminal four amino acids of BiP are Ala-Asp-Glu-Leu, a new variant of the XDEL sequence found at the C-termini of luminal endoplasmic reticulum proteins. To determine whether this sequence acts as a sorting signal in S. pombe we expressed an acid phosphatase ***fusion*** protein extended at its C-terminus with the amino acids ADEL. Analysis of the sorting of this ***fusion*** protein indicates that the ADEL sequence is sufficient to cause the retention of proteins in the endoplasmic reticulum. The sequences DDEL, HDEL and ***KDEL*** can also direct ER-retention of acid phosphatase in S. pombe.

L41 ANSWER 84 OF 92 EMBASE COPYRIGHT 2001 ELSEVIER SCI. B.V.
AN 92248020 EMBASE
DN 1992248020
TI In vitro effects of a recombinant toxin targeted to the fibroblast growth factor receptor on rat vascular smooth muscle and endothelial cells.
AU Biro S.; Siegall C.B.; Fu Y.-M.; Speir E.; Pastan I.; Epstein S.E.
CS Division of Cancer Biology, National Cancer Institute, National Institutes of Health,Bethesda, MD, United States
SO Circulation Research, (1992) 71/3 (640-645).
ISSN: 0009-7330 CODEN: CIRUAL
CY United States
DT Journal; Article
FS 005 General Pathology and Pathological Anatomy
029 Clinical Biochemistry
052 Toxicology
LA English
SL English
AB The dominant mechanism responsible for restenosis after angioplasty is believed to be the activation of medial smooth muscle cells (SMCs), leading to their proliferation, migration to the subintima, and further proliferation. To develop novel strategies that might inhibit or prevent restenosis, we previously used a ***chimeric*** toxin composed of transforming growth factor-.alpha. (which targets the epidermal growth factor receptor) and mutated Pseudomonas exotoxin to preferentially recognize and kill rapidly proliferating, versus quiescent, vascular SMCs. We have recently cloned and expressed a recombinant gene encoding Pseudomonas exotoxin with a mutated (nonfunctional) cell recognition domain fused with the ligand acidic fibroblast growth factor, termed aFGF-PE66(4Glu) ***KDEL*** ; thus, this recombinant toxin targets the fibroblast growth factor receptor. In the present study, we evaluated the relative effects of this ***chimeric*** toxin on quiescent versus rapidly proliferating vascular SMCs and also determined whether aFGF-PE66(4Glu) ***KDEL*** exerted different effects on SMCs versus endothelial cells. Rapidly proliferating SMCs (grown in 10% fetal bovine serum) were very sensitive to the cytotoxic effects of aFGF-PE66(4Glu) ***KDEL*** , whereas cytotoxicity was significantly less when the SMCs were in a quiescent state (grown in medium supplemented with 0.5% fetal bovine serum). The ***chimeric*** toxin was also significantly less cytotoxic against endothelial cells. Competition studies using excess acidic fibroblast growth factor indicated that the cytotoxic effects are specifically mediated by the fibroblast growth factor receptor. Thus, the present studies suggest a potentially expanded role of recombinant toxin therapy in restenosis: multiple receptors can be targeted, and cytotoxic effects, at least in vitro, can be preferentially directed to rapidly proliferating vascular SMCs, with relative sparing of vascular endothelial cells. It will next be necessary to test this strategy for inhibiting restenosis in an in vivo model of vascular injury and SMC proliferation.

L41 ANSWER 85 OF 92 CAPLUS COPYRIGHT 2001 ACS
AN 1992:486840 CAPLUS
DN 117:86840
TI Vicilin with carboxy-terminal ***KDEL*** is retained in the endoplasmic reticulum and accumulates to high levels in the leaves of transgenic plants
AU Wandelt, Christine I.; Khan, M. Rafiqul I.; Craig, Stuart; Schroeder, Harmut E.; Spencer, Donald; Higgins, Thomas J. V.
CS Div. Plant Ind., CSIRO, Canberra, 2601, Australia
SO Plant J. (1992), 2(2), 181-92
CODEN: PLJUED
DT Journal
LA English
AB Gene constructs were designed to test the effect of the endoplasmic reticulum (ER)-targeting signal, ***KDEL*** , on the level of accumulation of a foreign protein in transgenic plants. The gene for the pea seed protein vicilin was modified by the addn. of a sequence coding for this tetrapeptide at its carboxyl terminus. The altered gene was placed under the control of a CaMV 35S promoter and its expression in the leaves of both tobacco and lucerne (alfalfa) was compared with that of an equiv. vicilin construct lacking the ***KDEL*** -coding sequence. The

presence of the ER-targeting signal led to a greatly enhanced accumulation of the ***heterologous*** protein. In lucerne and tobacco leaves, the level of vicilin- ***KDEL*** protein was 20 and 100 times greater than that of the unmodified vicilin, resp. These differences in expression level could not be explained by corresponding differences in the steady-state levels or the translatability of the mRNAs. However, when the stability of vicilin and vicilin- ***KDEL*** proteins was compared with their resp. transgenic hosts, unmodified vicilin was found to be degraded with a half-life of 4.5 h while vicilin- ***KDEL*** was much more stable with a half-life of more than 48 h. Immunogold labeling of leaf tissues from transgenic lucerne and tobacco showed the presence of vicilin assocd. with large aggregates within the ER lumen of vicilin- ***KDEL*** plants. No such aggregates were detected in transgenic plants expressing wild-type vicilin. It is concluded that the carboxy-terminal ***KDEL*** caused the retention of the modified vicilin in the ER, and that this retention led to the increased stability and higher level of accumulation of vicilin- ***KDEL*** in leaves of transgenic plants.

L41 ANSWER 86 OF 92 CAPLUS COPYRIGHT 2001 ACS
AN 1991:466216 CAPLUS
DN 115:66216
TI Cytotoxicity regions of Pseudomonas exotoxin A and their use in immunotoxins
IN Pastan, I.; Chaudhary, V. K.; Fitzgerald, D.
PA National Institutes of Health, USA
SO U. S. Pat. Appl., 38 pp. Avail. NTIS Order No. PAT-APPL-6-522 563.
CODEN: XAXXAV
DT Patent
LA English
FAN.CNT 2

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI US 522563	A0	19910515	US 1990-522563	19900514
US 5458878	A	19951017		
US 459635	A0	19900415	US 1990-459635	19900102
WO 9118099	A1	19911128	WO 1991-US3121	19910510
W: AU, CA, JP				
RW: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LU, NL, SE				
AU 9179868	A1	19911210	AU 1991-79868	19910510
AU 660616	B2	19950706		
JP 05508537	T2	19931202	JP 1991-509974	19910510
EP 597844	A1	19940525	EP 1991-910455	19910510
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE				
CA 2082824	C	19981006	CA 1991-2082824	19910510
US 5705163	A	19980106	US 1995-461233	19950605
PRAI US 1990-459635		19900102		
US 1990-522563	A	19900514		
WO 1991-US3121	A	19910510		

AB Exotoxin A of Pseudomonas is modified to study the role of the C-terminal region in cytotoxicity and immunotoxins prepd. by addn. of appropriate ligand peptides to N-terminal, C-terminal, or both. Different ligands may be attached to the N- and C-termini to greatly increase the specificity of the toxin. C-terminal deletion analogs of exotoxin a were prepd. by expression of the cloned gene in Escherichia coli and the proteins tested for cytotoxicity and ADP-ribosylation activity. Cytotoxicity was completely lost when anything further than the last amino acid (position 613) was deleted. Deletion beyond amino acid 590 resulted in the loss of ADP-ribosylation activity. A pos. charged amino acid was found to be necessary at position 609, neg.-charged amino acids at positions 610 and 611 and leucine at position 612. When the C-terminal sequence REDLK was replaced with ***KDEL*** the protein was more cytotoxic. Addn. of two or three ***KDEL*** sequences further increased cytotoxicity. The prepn. of a bivalent toxin with ligands for binding cells carrying epidermal growth factor or interleukin-2, or both, was demonstrated.

L41 ANSWER 87 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 58
AN 1991:506007 BIOSIS
DN BA92:128967
TI INCREASED CYTOTOXIC ACTIVITY OF PSEUDOMONAS EXOTOXIN AND TWO
CHIMERIC TOXINS ENDING IN ***KDEL*** .
AU SEETHARAM S; CHAUDHARY V K; FITZGERALD D; PASTAN I
CS LABORATORY MOLECULAR BIOLOGY, DIVISION CANCER BIOLOGY, DIAGNOSIS CENTERS,
NCI, NIH, BUILD. 37, ROOM 4E16, BETHESDA, MD. 20892.
SO J BIOL CHEM, (1991) 266 (26), 17376-17381.
CODEN: JBCHA3. ISSN: 0021-9258.
FS BA; OLD
LA English
AB Pseudomonas exotoxin (PE) is a 66,000 molecular weight protein secreted by Pseudomonas aeruginosa. PE is made up of three domains, and PE40 is a form of PE which lacks domain Ia (amino acids 1-252) and has very low cytotoxicity because it cannot bind to target cells. The sequence Arg-Glu-Asp-Leu-Lys (REDLK) at the carboxyl terminus of Pseudomonas exotoxin has been shown to be important for its cytotoxic activity (Chaudhary, V. K., Jinno, Y., FitzGerald, D. J., and Pastan, I. (1990) Proc. Natl. Acad. Sci. U.S.A. 87, 308-312). In this study, we tested the effect of altering the carboxyl sequence of PE from REDLK to the characteristic endoplasmic reticulum retention sequence, ***KDEL*** , or to ***KDEL*** repeated three times (***KDEL***)3. We also made similar changes at the carboxyl terminus of two ***chimeric*** toxins in which domain I of PE (amino acids 1-252) was either replaced with transforming growth factor .alpha. (TGF.alpha.) to make TGF.alpha.-PE40 or with a single chain antibody (anti-Tac) reacting with the human interleukin 2 receptor to make anti-Tac(Fv)-PE40. Statistical analyses of our results demonstrate that PE and its derivatives ending in ***KDEL*** or (***KDEL***)3 are significantly more active than PE or derivatives ending in REDLK. We have also found that brefeldin A, which is known to perturb the endoplasmic reticulum, inhibits the cytotoxic action of PE. Our results suggest that the altered carboxyl terminus may enable the toxin to interact more efficiently with a cellular component involved in translocation of the toxin to the cytosol.

L41 ANSWER 88 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 59
AN 1992:2440 BIOSIS
DN BA93:2440
TI REGULATION OF EXPRESSION AND INTRACELLULAR DISTRIBUTION OF CALRETICULIN A
MAJOR CALCIUM BINDING PROTEIN OF NONMUSCLE CELLS.
AU OPAS M; DZIAK E; FLIEGEL L; MICHALAK M
CS DEP. ANAT., UNIV. TORONTO, TORONTO, ONTARIO, CAN. M5S 1A8.
SO J CELL PHYSIOL, (1991) 149 (1), 160-171.
CODEN: JCLLAX. ISSN: 0021-9541.
FS BA; OLD
LA English
AB In the present study we have demonstrated the presence of calreticulin, a major Ca2+-sequestering protein of nonmuscle cells, in a variety of cell types in tissue culture. The protein localizes to the endoplasmic reticulum in most cell types and also to the nuclear envelope or nucleoli-like structures in some cell types. Calreticulin is enriched in the rough endoplasmic reticulum, suggesting a possible involvement in protein synthesis. Calreticulin terminates with the ***KDEL*** -COOH sequence, which is likely responsible for its endoplasmic reticulum localization. Unlike some other ***KDEL*** proteins, calreticulin expression is neither heat-shock nor Ca2+-shock dependent. Using a variety of metabolic inhibitors, we have shown that the pool of calreticulin in L6 cells has a relatively slow turnover and a stable intracellular distribution. In proliferating muscle cells in culture (both L6 and human skeletal muscle) calreticulin is present in the endoplasmic reticulum, and additional intranuclear staining is observed. When ***fusion*** of the L6 cells is inhibited with either a high serum concentration or TGF-.beta. or TPA, the nucleolar staining by anticalreticulin antibodies is diminished, although the presence of calreticulin in the endoplasmic reticulum remains unchanged. In contrast, in differentiated (i.e., fused) muscle cells neither intranuclear nor intracellular staining for calreticulin is present. We conclude, therefore, that calreticulin is abundant in the endoplasmic reticulum in proliferating myoblasts, while it is present in only small amounts in sarcoplasmic reticulum membranes in terminally differentiated myotubes. We propose a model for the domain structure of calreticulin that may explain the differential subcellular distribution of this protein. Because of its widespread distribution in nonmuscle tissues, we postulate that calreticulin is a multifunctional protein that plays an important role in Ca2+ sequestering and thus that it is the nonmuscle analog of calsequestrin.

L41 ANSWER 89 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 60
AN 1992:25269 BIOSIS
DN BA93:14544
TI RETINOL-BINDING PROTEIN AND TRANSTHYRETIN EXPRESSED IN HELA CELLS FORM A
COMPLEX IN THE ENDOPLASMIC RETICULUM IN BOTH THE ABSENCE AND THE PRESENCE
OF RETINOL.
AU MELHUS H; NILSSON T; PETERSON P A; RASK L
CS DEP. CELL RES., UPPSALA UNIV., SWEDISH UNIV. AGRIC. SCI., UPPSALA BIOMED.
CENT., BOX 596, S-751 24 UPPSALA, SWED.
SO EXP CELL RES, (1991) 197 (1), 119-124.
CODEN: ECREAL. ISSN: 0014-4827.
FS BA; OLD
LA English
AB TO establish a suitable experimental system for studies of the interaction of retinol-binding protein (RBP) with transthyretin (TTR) we have expressed the corresponding cDNAs in HeLa cells. To investigate whether complex formation might occur already in the endoplasmic reticulum (ER), the C-terminal ER retention signal, ***KDEL*** , was attached to TTR. The tetrameric TTR- ***KDEL*** ***fusion*** protein was retained in the ER of HeLA cells. When RBP was co-expressed with TTR- ***KDEL*** , RBP was retained intracellularly. A cDNA-encoding purpurin, a protein which is 50% identical to RBP, was then expressed together with TTR- ***KDEL*** . Purpurin was not retained intracellularly and did not bind to TTR coupled to Sepharose. The effect of the vitamin A status on the secretion of TTR and RBP was examined. While TTR expressed alone was not retained intracellularly, TTR was retained in vitamin A deficient cells when co-expressed with RBP. Addition of retinol stimulated rapid secretion of both proteins. These results demonstrate that TTR can form a complex with RBP in the ER. The data suggest that RBP and TTR are secreted as a complex.

L41 ANSWER 90 OF 92 CAPLUS COPYRIGHT 2001 ACS
AN 1991:96250 CAPLUS
DN 114:96250
TI Cytotoxic recombinant Pseudomonas endotoxin and target-specific ***fusion*** products
IN Pastan, I.

PA National Institutes of Health, USA
SO U. S. Pat. Appl., 33 pp. Avail. NTIS Order No. PAT-APPL-7-759 635.
CODEN: XAXXAV
DT Patent
LA English
FAN.CNT 2

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI US 459635	A0	19900415	US 1990-459635	19900102
US 522563	A0	19910515	US 1990-522563	19900514
US 5458878	A	19951017		
CA 2072891	AA	19910703	CA 1990-2072891	19901227
WO 9109949	A1	19910711	WO 1990-US7421	19901227
W: AU, CA, JP				
RW: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LU, NL, SE				
AU 9172424	A1	19910724	AU 1991-72424	19901227
AU 644139	B2	19931202		
EP 509056	A1	19921021	EP 1991-904103	19901227
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE				
JP 05502032	T2	19930415	JP 1991-504333	19911217
US 5705163	A	19980106	US 1995-461233	19950605
PRAI US 1990-459635		19900102		
US 1990-522563	A3	19900514		
WO 1990-US7421	A	19901227		

AB The carboxyl terminus of Pseudomonas exotoxin A (PE), residues Arg609-Lys613, dets. the cytotoxic activity of the exotoxin. Peptide sequence Lys-Asp-Glu-Leu (***KDEL***), which is responsible for retaining newly formed proteins within the endoplasmic reticulum, has similar biol. function to the carboxyl terminus of PE. When ***KDEL*** is fused to a carboxyl terminus-deleted PE mutant (non-cytotoxic), it restored the cytotoxic activity of the toxin. A recognition mol. such as antibody may be fused to the carboxyl terminus of PE to increase the potency of the ***chimeric*** toxin. ***Fusion*** proteins of PE and transforming growth factor .alpha. were prepd., and their cytotoxic activity against Swiss 3T3 cells detd. The ***fusion*** proteins with active carboxyl terminus were .gtoreq.50 fold more cytotoxic than that contg. inactive PE carboxyl terminus.

L41 ANSWER 91 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 61
AN 1989:472262 BIOSIS
DN BA88:108022
TI COMPLEMENTARY DNA CLONES OF THE AUXIN-BINDING PROTEIN FROM CORN
COLEOPTILES ZEA-MAYS L. ISOLATION AND CHARACTERIZATION BY IMMUNOLOGICAL
METHODS.
AU TILLMANN U; VIOLA G; KAYSER B; SIEMEISTER G; HESSE T; PALME K; LEOBLER M;
KLAEMBT D
CS NATL. INST. HEALTH, BUILD. 6, ROOM 338, BETHESDA, MD. 20892, USA.
SO EMBO (EUR MOL BIOL ORGAN) J, (1989) 8 (9), 2463-2468.
CODEN: EMJODG. ISSN: 0261-4189.
FS BA; OLD
LA English
AB An auxin-binding protein (ABP) cDNA clone was selected from a .lambda.gt11 cDNA library from corn coleoptiles with highly purified IgGanti ABP. The sequence of 794 bp contains an open reading frame (ORF) of 603 bp, coding for a 22 kd protein. There are indications of a signal peptide of 38 amino acids (von Heijne, G. 1983, Eur. J. Biochem., 133, 17-21). A N-glycosylation site can be deduced and a C-terminal ***KDEL*** amino acid sequence is detected. An EcoRI fragment containing the beginning portion of the cDNA with about three quarters of the ORF was used to select cDNA clones from an independently produced .lambda.gt11 cDNA library of corn coleoptiles. Northern blot analysis with in vivo transcribed biotinylated RNA showed a single band of not more than 850 bases. The full-length in vitro transcript directed the in vitro synthesis of a protein which is precipitated by IgGanti ABP. Rabbit antibodies raised against a ***fusion*** protein detect the ABP as a double band on Western blots. Only the smaller of the two ABP bands is labeled by two different ***KDEL*** -specific IgG preparations.

L41 ANSWER 92 OF 92 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 62
AN 1989:94018 BIOSIS
DN BA87:48154
TI SORTING OF SOLUBLE ER PROTEINS IN YEAST.
AU PELHAM H R B; HARDWICK K G; LEWIS M J
CS MRC LAB. MOL. BIOL., HILLS ROAD, CAMBRIDGE CB2 2QH, UK.
SO EMBO (EUR MOL BIOL ORGAN) J, (1988) 7 (6), 1757-1762.
CODEN: EMJODG. ISSN: 0261-4189.
FS BA; OLD
LA English
AB In animal cells, luminal endoplasmic reticulum (ER) proteins are prevented from being secreted by a sorting system that recognizes the C-terminal sequence ***KDEL*** . We show that yeast has a similar sorting system, but it recognizes HDEL, rather than ***KDEL*** : derivatives of the enzyme invertase that bear the HDEL signal fail to be secreted. An invertase ***fusion*** protein that is retained in the cells is partially modified by outer-chain mannosyl transferase, which reside in the Golgi element. This supports the view, based on studies in animal cells, that ER targeting is achieved by continuous retrieval of proteins from the Golgi. We have used an invertase ***fusion*** gene to screen for mutants that are defective in this sorting system. Over 60 mutants were obtained; eight of these are alleles of a single gene, erd1. The mutant strains grow normally at 30.degree. C, but instead of retaining the ***fusion*** protein in the cells, they secrete it.

=> d his

(FILE 'HOME' ENTERED AT 09:34:11 ON 18 SEP 2001)

FILE 'BIOSIS, EMBASE, CAPLUS' ENTERED AT 09:34:29 ON 18 SEP 2001
L1 0 S LDLR354
L2 0 S LDLR 354
L3 1661 S LDLR
L4 879 S KDEL
L5 910 S KEEL
L6 263 S HDEL
L7 78 S DDEL
L8 9 S QDEL
L9 59 S ADEL
L10 16 S SDEL
L11 571052 S FUSION OR CHIMERIC OR HYBRID OR HETEROLOGOUS
L12 0 S L3 AND L4 AND L11
L13 0 S L3 AND L4
L14 0 S L3 AND L5 AND L11
L15 0 S L3 AND L6 AND L11
L16 0 S L3 AND L7
L17 0 S L3 AND L8
L18 0 S L3 AND L9
L19 0 S L3 AND L10
L20 81 S L3 AND L11
L21 195 S L4 AND L11
L22 40 S L5 AND L11
L23 60 S L6 AND L11
L24 3 S L7 AND L11
L25 3 S L8 AND L11
L26 5 S L9 AND L11
L27 1 S L10 AND L11
L28 34 DUP REM L20 (47 DUPLICATES REMOVED)
L29 9513 S LOW DENSITY LIPOPROTEIN RECEPTOR OR LDLR OR (LDLR AND 354)
L30 3 S L29 AND L4
L31 2 DUP REM L30 (1 DUPLICATE REMOVED)

FILE 'STNGUIDE' ENTERED AT 09:47:48 ON 18 SEP 2001

FILE 'BIOSIS, CAPLUS, EMBASE' ENTERED AT 09:48:26 ON 18 SEP 2001
L32 0 S L29 AND L5 AND L11
L33 0 S L29 AND L5
L34 0 S L29 AND L6
L35 0 S L29 AND L7
L36 0 S L29 AND L8
L37 0 S L29 AND L9
L38 0 S L29 AND L10
L39 322 S L29 AND L11
L40 168 DUP REM L39 (154 DUPLICATES REMOVED)
L41 92 DUP REM L21 (103 DUPLICATES REMOVED)
L42 34 DUP REM L22 (6 DUPLICATES REMOVED)
L43 25 DUP REM L23 (35 DUPLICATES REMOVED)
L44 1 DUP REM L24 (2 DUPLICATES REMOVED)
L45 1 DUP REM L25 (2 DUPLICATES REMOVED)
L46 3 DUP REM L26 (2 DUPLICATES REMOVED)
L47 1 DUP REM L27 (0 DUPLICATES REMOVED)

=> d bib abs l40

L40 ANSWER 1 OF 168 BIOSIS COPYRIGHT 2001 BIOSIS DUPLICATE 1
AN 2001:354346 BIOSIS
DN PREV200100354346
TI Differential functions of members of the ***low*** ***density*** ***lipoprotein*** ***receptor*** family suggested by their distinct endocytosis rates.
AU Li, Yonghe; Lu, Wenyan; Marzolo, Maria Paz; Bu, Guojun (1)
CS (1) Dept. of Pediatrics, Washington University School of Medicine, 660 South Euclid Ave., St. Louis, MO, 63110: bu@kids.wustl.edu USA
SO Journal of Biological Chemistry, (May 25, 2001) Vol. 276, No. 21, pp. 18000-18006. print.
ISSN: 0021-9258.
DT Article
LA English
SL English
AB The ***low*** ***density*** ***lipoprotein*** ***receptor*** (***LDLR***) family is composed of a class of cell surface endocytic receptors that recognize extracellular ligands and internalize them for degradation by lysosomes. In addition to ***LDLR*** , mammalian members of this family include the ***LDLR*** -related protein (LRP), the very ***low*** ***density*** ***lipoprotein*** ***receptor*** (VLDLR), the alipoprotein E receptor-2 (apoER2), and megalin. Herein we have analyzed the endocytic functions of the cytoplasmic tails of these receptors using LRP minireceptors, its ***chimeric*** receptor constructs, and full-length VLDLR and apoER2 stably expresseed in LRP-null Chinese hamster ovary cells. We find that the initial endocytosis rates mediated by different cytoplasmic tails are significantly different, with half-times of ligand internalization ranging from less than 30 s to more than 8 min. The tail of LRP mediates the highest rate of endocytosis,

whereas those of the VLDLR and apoER2 exhibit least endocytosis function. Compared with the tail of LRP, the tails of the ***LDLR*** and megalin display significantly lower levels of endocytosis rates. Ligand degradation analyses strongly support differential endocytosis rates initiated by these receptors. Interestingly, apoER2, which has recently been shown to mediate intracellular signal transduction, exhibited the lowest level of ligand degradation efficiency. These results thus suggest that the endocytic functions of members of the ***LDLR*** family are distinct and that certain receptors in this family may play their main roles in areas other than receptor-mediated endocytosis.

=> d bib abs 2-
1 ANSWERS ARE AVAILABLE. SPECIFIED ANSWER NUMBER EXCEEDS ANSWER SET SIZE
The answer numbers requested are not in the answer set.
ENTER ANSWER NUMBER OR RANGE (1):l40 2-
ANSWER NUMBERS NOT CORRECTLY SPECIFIED
Enter an answer number, Example: 10
several answer numbers, Example: 3,7,10
a range of answer numbers, Example: 5-10
or a combination of these. Example: 3,7,9-10,15
ENTER ANSWER NUMBER OR RANGE (1):
ENTER ANSWER NUMBER OR RANGE (1):l40
ANSWER NUMBERS NOT CORRECTLY SPECIFIED
Enter an answer number, Example: 10
several answer numbers, Example: 3,7,10
a range of answer numbers, Example: 5-10
or a combination of these. Example: 3,7,9-10,15
ENTER ANSWER NUMBER OR RANGE (1):2
1 ANSWERS ARE AVAILABLE. SPECIFIED ANSWER NUMBER EXCEEDS ANSWER SET SIZE
The answer numbers requested are not in the answer set.
ENTER ANSWER NUMBER OR RANGE (1):2,3
1 ANSWERS ARE AVAILABLE. SPECIFIED ANSWER NUMBER EXCEEDS ANSWER SET SIZE
The answer numbers requested are not in the answer set.
ENTER ANSWER NUMBER OR RANGE (1):
ENTER ANSWER NUMBER OR RANGE (1):
ENTER ANSWER NUMBER OR RANGE (1):

ENTER ANSWER NUMBER OR RANGE (1):

ENTER ANSWER NUMBER OR RANGE (1):

YOU HAVE RECEIVED THIS PROMPT MESSAGE 5 CONSECUTIVE TIMES WITHOUT ENTERING A REQUESTED RESPONSE
Enter an answer number, Example: 10
several answer numbers, Example: 3,7,10
a range of answer numbers, Example: 5-10
or a combination of these. Example: 3,7,9-10,15
IF YOU REQUIRE FURTHER HELP, PLEASE CONTACT YOUR LOCAL HELP DESK
ENTER ANSWER NUMBER OR RANGE (1):

ENTER ANSWER NUMBER OR RANGE (1):
ENTER ANSWER NUMBER OR RANGE (1):
ENTER ANSWER NUMBER OR RANGE (1):
ENTER ANSWER NUMBER OR RANGE (1):

---Logging off of STN---

ENTER ANSWER NUMBER OR RANGE (1):END

=>
Executing the logoff script...

=> LOG Y

COST IN U.S. DOLLARS	ENTRY	SINCE FILE SESSION	TOTAL
FULL ESTIMATED COST		333.43	388.02

DISCOUNT AMOUNTS (FOR QUALIFYING ACCOUNTS)	ENTRY	SESSION	SINCE FILE TOTAL
CA SUBSCRIBER PRICE		-15.29	-15.29

****

Dear valued customer,

Your feedback is important to us. Would you kindly take a moment to complete our survey? This survey will only take about 5-10 minutes to complete. Your responses will be kept confidential and will help us improve STN Express with Discover! for your future use. Please click on the following link to access the survey.

http://www.cas.org/ONLINE/STN/ExpressSurveyForm.html?LOGINID=SSSPTA1633CXQ

****

STN INTERNATIONAL LOGOFF AT 10:09:33 ON 18 SEP 2001

Trying 3106016892...Open

Welcome to STN International! Enter x:x
LOGINID:ssspta1633cxq
PASSWORD:
****** RECONNECTED TO STN INTERNATIONAL ******
SESSION RESUMED IN FILE 'BIOSIS, CAPLUS, EMBASE' AT 10:10:18 ON 18 SEP 2001
FILE 'BIOSIS' ENTERED AT 10:10:18 ON 18 SEP 2001
COPYRIGHT (C) 2001 BIOSIS(R)
FILE 'CAPLUS' ENTERED AT 10:10:18 ON 18 SEP 2001
COPYRIGHT (C) 2001 AMERICAN CHEMICAL SOCIETY (ACS)
FILE 'EMBASE' ENTERED AT 10:10:18 ON 18 SEP 2001
COPYRIGHT (C) 2001 Elsevier Science B.V. All rights reserved.
STN INTERNATIONAL LOGOFF AT 10:09:33 ON 18 SEP 2001

---Logging off of STN---

END

Unable to generate the STN prompt.
Exiting the script...
Trying 3106016892...Open

Welcome to STN International! Enter x:x
LOGINID:ssspta1633cxq
PASSWORD:
TERMINAL (ENTER 1, 2, 3, OR ?):2

********** Welcome to STN International **********

NEWS 1 Web Page URLs for STN Seminar Schedule - N. America
NEWS 2 Dec 17 The CA Lexicon available in the CAPLUS and CA files
NEWS 3 Feb 06 Engineering Information Encompass files have new names
NEWS 4 Feb 16 TOXLINE no longer being updated
NEWS 5 Apr 23 Search Derwent WPINDEX by chemical structure
NEWS 6 Apr 23 PRE-1967 REFERENCES NOW SEARCHABLE IN CAPLUS AND CA
NEWS 7 May 07 DGENE Reload
NEWS 8 Jun 20 Published patent applications (A1) are now in USPATFULL
NEWS 9 JUL 13 New SDI alert frequency now available in Derwent's DWPI and DPCI
NEWS 10 Aug 23 In-process records and more frequent updates now in MEDLINE
NEWS 11 Aug 23 PAGE IMAGES FOR 1947-1966 RECORDS IN CAPLUS AND CA
NEWS 12 Aug 23 Adis Newsletters (ADISNEWS) now available on STN
NEWS 13 Sep 17 IMSworld Pharmaceutical Company Directory name change to PHARMASEARCH

NEWS EXPRESS August 15 CURRENT WINDOWS VERSION IS V6.0c, CURRENT MACINTOSH VERSION IS V6.0 (ENG) AND V6.0J (JP), AND CURRENT DISCOVER FILE IS DATED 07 AUGUST 2001
NEWS HOURS STN Operating Hours Plus Help Desk Availability
NEWS INTER General Internet Information
NEWS LOGIN Welcome Banner and News Items
NEWS PHONE Direct Dial and Telecommunication Network Access to STN
NEWS WWW CAS World Wide Web Site (general information)

Enter NEWS followed by the item number or name to see news on that specific topic.

All use of STN is subject to the provisions of the STN Customer agreement. Please note that this agreement limits use to scientific research. Use for software development or design or implementation of commercial gateways or other similar uses is prohibited and may result in loss of user privileges and other penalties.

********** STN Columbus **********

FILE 'HOME' ENTERED AT 10:11:58 ON 18 SEP 2001

=> file medline caplus biosis

COST IN U.S. DOLLARS	ENTRY	SINCE FILE SESSION	TOTAL
FULL ESTIMATED COST		0.15	0.15

FILE 'MEDLINE' ENTERED AT 10:12:16 ON 18 SEP 2001

FILE 'CAPLUS' ENTERED AT 10:12:16 ON 18 SEP 2001
USE IS SUBJECT TO THE TERMS OF YOUR STN CUSTOMER AGREEMENT.
PLEASE SEE "HELP USAGETERMS" FOR DETAILS.
COPYRIGHT (C) 2001 AMERICAN CHEMICAL SOCIETY (ACS)

FILE 'BIOSIS' ENTERED AT 10:12:16 ON 18 SEP 2001
COPYRIGHT (C) 2001 BIOSIS(R)

=> s low density lipoprotein receptor or LDLR or (LDLR and 354)
L1 7533 LOW DENSITY LIPOPROTEIN RECEPTOR OR LDLR OR (LDLR AND 354)

=> s KDEL
L2 931 KDEL

=> s l1 and l2
L3 2 L1 AND L2

=> s l1 and (fusion or chimeric or hybrid or heterologous)
L4 359 L1 AND (FUSION OR CHIMERIC OR HYBRID OR HETEROLOGOUS)

=> s l2 and (fusion or chimeric or hybrid or heterologous)
L5 229 L2 AND (FUSION OR CHIMERIC OR HYBRID OR HETEROLOGOUS)

=> dup rem l3
PROCESSING COMPLETED FOR L3
L6 2 DUP REM L3 (0 DUPLICATES REMOVED)

=> dup rem l4
PROCESSING COMPLETED FOR L4
L7 206 DUP REM L4 (153 DUPLICATES REMOVED)

=> dup rem l5
PROCESSING COMPLETED FOR L5
L8 113 DUP REM L5 (116 DUPLICATES REMOVED)

=> d bib abs l8 1-
YOU HAVE REQUESTED DATA FROM 113 ANSWERS - CONTINUE? Y/(N):y

L8 ANSWER 1 OF 113 CAPLUS COPYRIGHT 2001 ACS
AN 2001:45049 CAPLUS
DN 134:97534
TI Conjugates for the delivery of active substances into cells, cell compartments and membranes
IN Braun, Klaus; Friedrich, Eckart; Waldeck, Waldemar; Peschke, Peter; Pipkorn, Ruediger; Debus, Juergen
PA Deutsches Krebsforschungszentrum Stiftung des Oeffentlichen Rechts, Germany
SO Ger. Offen., 10 pp.
CODEN: GWXXBX
DT Patent
LA German
FAN.CNT 1

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI DE 19933492	A1	20010118	DE 1999-19933492	19990716
WO 2001005432	A2	20010125	WO 2000-DE2346	20000714

W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CR, CU, CZ, DK, DM, DZ, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG

PRAI DE 1999-19933492 A 19990716
AB The invention concerns the prodn. and application of conjugates for the delivery of active substance into cells, cell compartments and membranes that contain fragments of a penetrating protein, a target-specific localization protein and the active substance. Cell-penetrating proteins are penetratin, transportan or their derivs. Sequences of the target-specific localization peptides are given for endoplasmic reticulum, mitochondria, nucleus, peroxisomes and cell membrane. Active substances are diagnostic agents or drugs. Spacers can be included into the conjugate between the active substance and the target-specific peptide. Synthesis methods include the Merrifield synthesis and coupling of the non-peptide component. Thus penetratin, a nuclear localization sequence and a spacer sequence peptide-conjugate was synthesized; after purifn., it was coupled with rhodamine 110. The conjugate was incubated with AT-1 and DU-145 cells; the penetration of the rhodamine 110 contg. conjugate into the nucleus was detected by fluoroscence microscopy.
RE.CNT 3
RE
(1) Anon; Drug Design 1980, VX, PS226
(2) Anon; Molecular Biology of the Cell 1983, PS344
(3) Anon; Rompp Chemie Lexikon 1998, V10, Ps2584

L8 ANSWER 2 OF 113 MEDLINE DUPLICATE 1
AN 2001403588 MEDLINE
DN 21347877 PubMed ID: 11323436
TI Ykt6 forms a SNARE complex with syntaxin 5, GS28, and Bet1 and participates in a late stage in endoplasmic reticulum-Golgi transport.
AU Zhang T; Hong W
CS Membrane Biology Laboratory, Institute of Molecular and Cell Biology, Singapore 117609, Singapore.
SO JOURNAL OF BIOLOGICAL CHEMISTRY, (2001 Jul 20) 276 (29) 27480-7.
Journal code: HIV; 2985121R. ISSN: 0021-9258.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 200108
ED Entered STN: 20010827
Last Updated on STN: 20010827
Entered Medline: 20010823
AB The yeast SNARE Ykt6p has been implicated in several trafficking steps, including vesicular transport from the endoplasmic reticulum (ER) to the Golgi, intra-Golgi transport, and homotypic vacuole ***fusion*** . The functional role of its mammalian homologue (Ykt6) has not been established. Using antibodies specific for mammalian Ykt6, it is revealed that it is found mainly in Golgi-enriched membranes. Three SNAREs, syntaxin 5, GS28, and Bet1, are specifically associated with Ykt6 as revealed by co-immunoprecipitation, suggesting that these four SNAREs form a SNARE complex. Double labeling of Ykt6 and the Golgi marker mannosidase II or the ER-Golgi recycling marker ***KDEL*** receptor suggests that Ykt6 is primarily associated with the Golgi apparatus. Unlike the ***KDEL*** receptor, Ykt6 does not cycle back to the peripheral ER exit sites. Antibodies against Ykt6 inhibit in vitro ER-Golgi transport of vesicular stomatitis virus envelope glycoprotein (VSVG) only when they are added before the EGTA-sensitive stage. ER-Golgi transport of VSVG in vitro is also inhibited by recombinant Ykt6. In the presence of antibodies against Ykt6, VSVG accumulates in peri-Golgi vesicular structures and is prevented from entering the mannosidase II compartment, suggesting that Ykt6 functions at a late stage in ER-Golgi transport. Golgi apparatus marked by mannosidase II is fragmented into vesicular structures in cells microinjected with Ykt6 antibodies. It is concluded that Ykt6 functions in a late step of ER-Golgi transport, and this role may be important for the integrity of the Golgi complex.

L8 ANSWER 3 OF 113 MEDLINE
AN 2001371655 MEDLINE
DN 21299367 PubMed ID: 11406585
TI The ***KDEL*** receptor mediates a retrieval mechanism that contributes to quality control at the endoplasmic reticulum.
AU Yamamoto K; Fujii R; Toyofuku Y; Saito T; Koseki H; Hsu V W; Aoe T
CS Department of Molecular Embryology, Chiba University Graduate School of Medicine, Chiba 260-8670, Japan.
SO EMBO JOURNAL, (2001 Jun 15) 20 (12) 3082-91.
Journal code: EMB; 8208664. ISSN: 0261-4189.
CY England: United Kingdom
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 200107
ED Entered STN: 20010730
Last Updated on STN: 20010730
Entered Medline: 20010726
AB Newly synthesized proteins in the endoplasmic reticulum (ER) must fold and assemble correctly before being transported to their final cellular destination. While some misfolded or partially assembled proteins have been shown to exit the ER, they fail to escape the early secretory system entirely, because they are retrieved from post-ER compartments to the ER. We elucidate a mechanistic basis for this retrieval and characterize its contribution to ER quality control by studying the fate of the unassembled T-cell antigen receptor (TCR) alpha chain. While the steady-state distribution of TCRalpha is in the ER, inhibition of retrograde transport by COPI induces the accumulation of TCRalpha in post-ER compartments, suggesting that TCRalpha is cycling between the ER and post-ER compartments. TCRalpha associates with BiP, a ***KDEL*** protein. Disruption of the ligand-binding function of the ***KDEL*** receptor releases TCRalpha from the early secretory system to the cell surface, so that TCRalpha is no longer subject to ER degradation. Thus, our findings suggest that retrieval by the ***KDEL*** receptor contributes to mechanisms by which the ER monitors newly synthesized proteins for their proper disposal.

L8 ANSWER 4 OF 113 MEDLINE DUPLICATE 2
AN 2001252506 MEDLINE
DN 21248724 PubMed ID: 11351308
TI Isolation of new anti-CD30 scFvs from DNA-immunized mice by phage display and biologic activity of recombinant immunotoxins produced by ***fusion*** with truncated pseudomonas exotoxin.
AU Rozemuller H; Chowdhury P S; Pastan I; Kreitman R J
CS Laboratory of Molecular Biology, National Cancer Institute, National Institutes of Health, Bethesda, MD, USA.
SO INTERNATIONAL JOURNAL OF CANCER, (2001 Jun 15) 92 (6) 861-70.
Journal code: GQU; 0042124. ISSN: 0020-7136.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals

EM 200106
ED Entered STN: 20010702
Last Updated on STN: 20010702
Entered Medline: 20010628
AB To target CD30 on Hodgkin's disease and anaplastic large-cell lymphoma, anti-CD30 single-chain antibodies were obtained by DNA immunization of mice with the complete human CD30 cDNA. Spleens were isolated from mice with high anti-CD30 titer, and the RNA was used for the production of an scFv-displaying phage library. Specific phages were enriched by 3 rounds of panning on soluble CD30 or CD30+ K562 cells. Recombinant immunotoxins (rITs) were made from 3 ELISA-positive scFv phages by ***fusion*** to a 38 kDa truncated mutant of Pseudomonas exotoxin (PE38) with or without a ***KDEL*** mutant sequence at the C terminus. In vitro cytotoxicity of purified anti-CD30 rITs was measured on CD30-transfected A431 cells. IC50 values ranged from 3 to 7 ng/ml (50-110 pM) for PE38 rITs and 0.1 ng/ml (2 pM) for the PE38- ***KDEL*** IT on A431-CD30 cells. The parental A431 cells were resistant, indicating that the cytotoxicity was specific and CD30-mediated. rITs were tested for anti-tumor activity in a nude mouse model. A431-CD30 cells were injected s.c. on day 0; then, mice bearing measurable tumors were treated beginning on day 4 with 3 alternate daily doses i.v. Anti-tumor activity was dose-dependent and not found when irrelevant ITs were administered or when CD30- tumors were treated. Our data show that DNA immunization and antibody phage display may be useful in producing new rITs against hematologic malignancies. Published 2001 Wiley-Liss, Inc.

L8 ANSWER 5 OF 113 MEDLINE DUPLICATE 3
AN 2001480600 MEDLINE
DN 21414648 PubMed ID: 11523796
TI Intracellular apolipoprotein E affects Amyloid Precursor Protein processing and amyloid Abeta production in COS-1 cells.
AU Hass S; Weidemann A; Utermann G; Baier G
CS Institute for Medical Biology and Human Genetics, University of Innsbruck, Austria.
SO Mol Genet Genomics, (2001 Jul) 265 (5) 791-800.
Journal code: D2D; 101093320. ISSN: 1617-4615.
CY Germany: Germany, Federal Republic of
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 200109
ED Entered STN: 20010830
Last Updated on STN: 20010917
Entered Medline: 20010913
AB The apoE gene has been identified as a major susceptibility locus for late-onset Alzheimer's disease (LOAD). The epsilon4 allele greatly reduces age of onset of LOAD as compared to the wild-type 3 allele. The molecular mechanism(s) underlying the association has not yet been fully elucidated. The apoE protein has been shown to physically interact with the Abeta region of the Amyloid Precursor Protein (APP), but also with the ectodomain of the APP holoprotein itself. In this study we have used apoE ***fusion*** proteins containing either the ER retention sequence ***KDEL*** or trans-Golgi network (TGN) signal sequence in order to define potential apoE-mediated alterations in APP protein processing. Co-expression and pulse-chase experiments showed that a functional apoE:APP interaction occurs intracellularly which directly affects maturation and subsequently the secretion kinetics of APP. In addition, an epsilon4 allele-specific induction of Abeta production has been demonstrated. apoE3 resulted in increased Abeta production only when targeted to the ER, as observed in cells transfected with an apoE3KDEL ***fusion*** protein as well as following treatment with brefeldin A. The findings suggest that in cells that express both apoE and APP, such as astrocytes and microglia, a functional apoE:APP interaction may occur which modulates APP processing and Abeta production.

L8 ANSWER 6 OF 113 MEDLINE
AN 2001443700 MEDLINE
DN 21382255 PubMed ID: 11489915
TI Head-to-tail oligomerization of calsequestrin: a novel mechanism for heterogeneous distribution of endoplasmic reticulum luminal proteins.
AU Gatti G; Trifari S; Mesaeli N; Parker J M; Michalak M; Meldolesi J
CS Department of Pharmacology, University of Milan, 20129 Milan, Italy.
SO JOURNAL OF CELL BIOLOGY, (2001 Aug 6) 154 (3) 525-34.
Journal code: HMV; 0375356. ISSN: 0021-9525.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 200109
ED Entered STN: 20010813
Last Updated on STN: 20010910
Entered Medline: 20010906
AB Many proteins retained within the endo/sarcoplasmic reticulum (ER/SR) lumen express the COOH-terminal tetrapeptide ***KDEL*** , by which they continuously recycle from the Golgi complex; however, others do not express the ***KDEL*** retrieval signal. Among the latter is calsequestrin (CSQ), the major Ca2+-binding protein condensed within both the terminal cisternae of striated muscle SR and the ER vacuolar domains of some neurons and smooth muscles. To reveal the mechanisms of condensation and establish whether it also accounts for ER/SR retention of CSQ, we generated a variety of constructs: chimeras with another similar protein, calreticulin (CRT); mutants truncated of COOH- or NH2-terminal domains; and other mutants deleted or point mutated at strategic sites. By transfection in L6 myoblasts and HeLa cells we show here that CSQ condensation in ER-derived vacuoles requires two amino acid sequences, one at the NH2 terminus, the other near the COOH terminus. Experiments with a green fluorescent protein GFP/CSQ chimera demonstrate that the CSQ-rich vacuoles are long-lived organelles, unaffected by Ca2+ depletion, whose almost complete lack of movement may depend on a direct interaction with the ER. CSQ retention within the ER can be dissociated from condensation, the first identified process by which ER luminal proteins assume a heterogeneous distribution. A model is proposed to explain this new process, that might also be valid for other luminal proteins.

L8 ANSWER 7 OF 113 CAPLUS COPYRIGHT 2001 ACS DUPLICATE 4
AN 2001:411325 CAPLUS
DN 135:87689
TI Expression of a sulphur-rich sunflower albumin gene in transgenic tall fescue (Festuca arundinacea Schreb.) plants
AU Wang, Z. Y.; Ye, X. D.; Nagel, J.; Potrykus, I.; Spangenberg, G.
CS Plant Biotechnology Centre, Agriculture Victoria and CRC for Molecular Plant Breeding, La Trobe University, Bundoora, 3083, Australia
SO Plant Cell Rep. (2001), 20(3), 213-219
CODEN: PCRPD8; ISSN: 0721-7714
PB Springer-Verlag
DT Journal
LA English
AB Transgenic tall fescue (Festuca arundinacea Schreb.) plants have been generated that express foreign genes encoding a rumen-stable protein rich in sulfur-contg. amino acids. The aim was to improve the protein quality of a forage grass for ruminant nutrition. ***Chimeric*** sulfur-rich sunflower albumin (SFA8) genes, including an endoplasmic reticulum retention signal (***KDEL***), were constructed under the control of constitutive (CaMV 35S) and light-regulated (wheat Cab) promoters. These constructs were introduced into the tall fescue genome by micro-projectile bombardment of embryogenic suspension cells. The sunflower albumin transgenes stably integrated into the plant genome as demonstrated by Southern hybridization anal. The transgenic tall fescue plants produced the expected transcript, and the corresponding sulfur-rich SFA8 protein accumulated up to 0.2% of the total sol. protein in individual transgenic plants.
RE.CNT 46
RE
(1) Asano, Y; Plant Cell Rep 1994, V13, P243 CAPLUS
(2) Barry, T; Br J Nutr 1981, V46, P521 CAPLUS
(3) Bilang, R; Gene 1991, V100, P247 CAPLUS
(4) Bradford, M; Anal Biochem 1976, V72, P248 CAPLUS
(6) Dalton, S; Plant Sci 1995, V108, P63 CAPLUS
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L8 ANSWER 8 OF 113 CAPLUS COPYRIGHT 2001 ACS
AN 2001:532883 CAPLUS
TI ***KDEL*** -cargo regulates interactions between proteins involved in COPI vesicle traffic: measurements in living cells using FRET
AU Majoul, Irina; Straub, Martin; Hell, Stefan W.; Duden, Rainer; Soling, Hans-Dieter
CS Department of Neurobiology, Max-Planck-Institute of Biophysical Chemistry, Gottingen, D-37077, Germany
SO Dev. Cell (2001), 1(1), 139-153
CODEN: DCEEBE; ISSN: 1534-5807
PB Cell Press
DT Journal
LA English
AB How the occupied ***KDEL*** receptor ERD2 is sorted into COPI vesicles for Golgi-to-ER transport is largely unknown. Here, interactions between proteins of the COPI transport machinery occurring during a "wave" of transport of a ***KDEL*** ligand were studied in living cells. FRET between CFP and YFP ***fusion*** proteins was measured by multifocal multiphoton microscopy and bulk-cell spectrofluorimetry. Ligand binding induces oligomerization of ERD2 and recruitment of ARFGAP to the Golgi, where the (ERD2)n/ARFGAP complex interacts with membrane-bound ARF1. During ***KDEL*** ligand transport, interactions of ERD2 with .beta.-COP and p23 decrease and the proteins segregate. Both p24a and p23 interact with ARF1, but only p24 interacts with ARFGAP. These findings suggest a model for how cargo-induced oligomerization of ERD2 regulates its sorting into COPI-coated buds.
RE.CNT 51
RE
(1) Aoe, T; EMBO J 1997, V16, P7305 CAPLUS
(2) Aoe, T; Proc Natl Acad Sci USA 1998, V95, P1624 CAPLUS
(3) Barlowe, C; Traffic 2000, V1, P371 CAPLUS
(5) Blum, R; J Cell Sci 1999, V112, P537 CAPLUS
(6) Bremser, M; Cell 1999, V96, P495 CAPLUS
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L8 ANSWER 9 OF 113 CAPLUS COPYRIGHT 2001 ACS
AN 2000:368420 CAPLUS
DN 133:3725
TI Suppression of xenotransplant rejection
IN Ramrakha, Punit Satyavrat; George, Andrew John Timothy; Haskard, Dorian; Lechler, Robert Ian
PA Imperial College Innovations Limited, UK
SO PCT Int. Appl., 36 pp.
CODEN: PIXXD2
DT Patent
LA English

FAN.CNT 1

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI WO 2000031126	A2	20000602	WO 1999-GB3888	19991122
WO 2000031126	A3	20000824		

W: AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CR, CU, CZ, DE, DK, DM, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM

RW: GH, GM, KE, LS, MW, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG

EP 1131411	A2	20010912	EP 1999-956179	19991122

R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO

PRAI GB 1998-25555	A	19981120		
WO 1999-GB3888	W	19991122		

AB The authors disclose methods for suppression of graft rejection, particularly xenograft rejection. In one example, a phage library was created for human antibodies directed to porcine VCAM. Phage-derived scFvs were engineered to express the endoplasmic reticulum targeting signal ***KDEL*** and transfected into porcine aortic endothelial cells. FACS anal. showed a redn. in VCAM surface expression and a functional loss in adhesive function as demonstrated by reduced binding to Jurkat cells.

L8 ANSWER 10 OF 113 CAPLUS COPYRIGHT 2001 ACS
AN 2000:98760 CAPLUS
DN 132:133894
TI Inhibition of ***KDEL*** receptor-mediated return of heat shock protein complexes to the endoplasmic reticulum and their adjuvant use
IN Rothman, James E.; Mayhew, Mark; Hoe, Mee H.
PA Sloan-Kettering Institute for Cancer Research, USA
SO PCT Int. Appl., 87 pp.
CODEN: PIXXD2
DT Patent
LA English
FAN.CNT 1

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI WO 2000006729	A1	20000210	WO 1999-US17147	19990728

W: AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM

RW: GH, GM, KE, LS, MW, SD, SL, SZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG

US 6160088	A	20001212	US 1998-124671	19980729
AU 9953245	A1	20000221	AU 1999-53245	19990728
EP 1100906	A1	20010523	EP 1999-938851	19990728

R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO

PRAI US 1998-124671	A	19980729		
WO 1999-US17147	W	19990728		

AB Inhibitors of the ***KDEL*** receptor that can be used to block the transfer of heat shock proteins to the endoplasmic reticulum and allow them to act as adjuvants are described. Certain proteins are functionally retained in the cellular endoplasmic reticulum via an interaction between a ***KDEL*** sequence and its receptor. According to the invention, blocking this interaction with a ***KDEL*** receptor inhibitor promotes the secretion of such proteins. In specific embodiments of the invention, ***KDEL*** receptor inhibitors may be used to promote the secretion of heat shock proteins, thereby rendering the secreted heat shock proteins more accessible to the immune system and improving the immune response to heat shock protein-assocd. antigens. The inhibitors are artificial peptides that oligomerize and present large no. of ***KDEL*** peptides to the receptors and sat. them. An example of one of these peptides uses the signal peptide of the BiP protein, an oligomerization domain of a cartilage oligomeric matrix protein, a linker peptide from a camel Ig and a ***KDEL*** peptide is described.

RE.CNT 2
RE
(1) Ciba Geigy Ag; WO 9818943 A 1998 CAPLUS
(2) Sloan-Kettering Institute For Cancer Research; WO 9706828 A 1997 CAPLUS

L8 ANSWER 11 OF 113 MEDLINE
AN 2000496175 MEDLINE
DN 20435885 PubMed ID: 10864930
TI Membrane recruitment of coatomer and binding to dilysine signals are separate events.
AU Gomez M; Scales S J; Kreis T E; Perez F
CS Department of Cell Biology, University of Geneva, Sciences III, 30 Quai Ernest-Ansermet, CH-1211 Geneva 4, Switzerland.
SO JOURNAL OF BIOLOGICAL CHEMISTRY, (2000 Sep 15) 275 (37) 29162-9.
Journal code: HIV; 2985121R. ISSN: 0021-9258.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 200010
ED Entered STN: 20001027
Last Updated on STN: 20001027
Entered Medline: 20001013

AB It has previously been shown that transport of newly synthesized proteins and the structure of the Golgi complex are affected in the Chinese hamster ovary cell line ldlF, which bears a temperature-sensitive mutation in the Coat protein I (COPI) subunit epsilon-COP (Guo, Q., Vasile, E., and Krieger, M. (1994) J. Cell Biol. 125, 1213-1224; Hobbie, L., Fisher, A. S., Lee, S., Flint, A., and Krieger, M. (1994) J. Biol. Chem. 269, 20958-20970). Here, we pinpoint the site of the secretory block to an intermediate compartment between the endoplasmic reticulum (ER) and the Golgi complex and show that the distributions of ER-Golgi recycling proteins, such as ***KDEL*** receptor and p23, as well as resident Golgi proteins, such as mannosidase II, are accordingly affected. At the nonpermissive temperature, neither the stability of the COPI complex nor its recruitment to donor Golgi membranes is affected. However, the binding of coatomer to the dilysine-based ER-retrieval motif is impaired in the absence of epsilon-COP, suggesting that dilysine signal binding is not the major means of COPI recruitment. Because expression of the exogenous chimera of epsilon-COP and green fluorescent protein in ldlF cells at nonpermissive temperature rapidly restores the wild type properties, epsilon-COP is likely to play an important role in the cargo selection events mediated by COPI.

L8 ANSWER 12 OF 113 MEDLINE DUPLICATE 5
AN 2000127874 MEDLINE
DN 20127874 PubMed ID: 10660554
TI Retention of subunits of the oligosaccharyltransferase complex in the endoplasmic reticulum.
AU Fu J; Kreibich G
CS Department of Cell Biology, New York University School of Medicine, New York, New York 10016, USA.
SO JOURNAL OF BIOLOGICAL CHEMISTRY, (2000 Feb 11) 275 (6) 3984-90.
Journal code: HIV; 2985121R. ISSN: 0021-9258.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 200003
ED Entered STN: 20000327
Last Updated on STN: 20000327
Entered Medline: 20000316

AB Membrane proteins of the endoplasmic reticulum (ER) may be localized to this organelle by mechanisms that involve retention, retrieval, or a combination of both. For luminal ER proteins, which contain a ***KDEL*** domain, and for type I transmembrane proteins carrying a dilysine motif, specific retrieval mechanisms have been identified. However, most ER membrane proteins do not contain easily identifiable retrieval motifs. ER localization information has been found in cytoplasmic, transmembrane, or luminal domains. In this study, we have identified ER localization domains within the three type I transmembrane proteins, ribophorin I (RI), ribophorin II (RII), and OST48. Together with DAD1, these membrane proteins form an oligomeric complex that has oligosaccharyltransferase (OST) activity. We have previously shown that ER retention information is independently contained within the transmembrane and the cytoplasmic domain of RII, and in the case of RI, a truncated form consisting of the luminal domain was retained in the ER. To determine whether other domains of RI carry additional retention information, we have generated chimeras by exchanging individual domains of the Tac antigen with the corresponding ones of RI. We demonstrate here that only the luminal domain of RI contains ER retention information. We also show that the dilysine motif in OST48 functions as an ER localization motif because OST48 in which the two lysine residues are replaced by serine (OST48ss) is no longer retained in the ER and is found instead also at the plasma membrane. OST48ss is, however, retained in the ER when coexpressed with RI, RII, or chimeras, which by themselves do not exit from the ER, indicating that they may form partial oligomeric complexes by interacting with the luminal domain of OST48. In the case of the Tac chimera containing only the luminal domain of RII, which by itself exits from the ER and is rapidly degraded, it is retained in the ER and becomes stabilized when coexpressed with OST48.

L8 ANSWER 13 OF 113 MEDLINE
AN 2001040260 MEDLINE
DN 20483777 PubMed ID: 11029049
TI Identification of a novel saturable endoplasmic reticulum localization mechanism mediated by the C-terminus of a Dictyostelium protein disulfide isomerase.
AU Monnat J; Neuhaus E M; Pop M S; Ferrari D M; Kramer B; Soldati T
CS Department of Molecular Cell Research, Max-Planck-Institute for Medical Research, D-69120 Heidelberg, Germany.
SO MOLECULAR BIOLOGY OF THE CELL, (2000 Oct) 11 (10) 3469-84.
Journal code: BAU. ISSN: 1059-1524.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 200012
ED Entered STN: 20010322
Last Updated on STN: 20010322
Entered Medline: 20001207

AB Localization of soluble endoplasmic reticulum (ER) resident proteins is

likely achieved by the complementary action of retrieval and retention mechanisms. Whereas the machinery involving the H/ ***KDEL*** and related retrieval signals in targeting escapees back to the ER is well characterized, other mechanisms including retention are still poorly understood. We have identified a protein disulfide isomerase (Dd-PDI) lacking the HDEL retrieval signal normally found at the C terminus of ER residents in Dictyostelium discoideum. Here we demonstrate that its 57 residue C-terminal domain is necessary for intracellular retention of Dd-PDI and sufficient to localize a green fluorescent protein (GFP) chimera to the ER, especially to the nuclear envelope. Dd-PDI and GFP-PDI57 are recovered in similar cation-dependent complexes. The overexpression of GFP-PDI57 leads to disruption of endogenous PDI complexes and induces the secretion of PDI, whereas overexpression of a GFP-HDEL chimera induces the secretion of endogenous calreticulin, revealing the presence of two independent and saturable mechanisms. Finally, low-level expression of Dd-PDI but not of PDI truncated of its 57 C-terminal residues complements the otherwise lethal yeast TRG1/PDI1 null mutation, demonstrating functional disulfide isomerase activity and ER localization. Altogether, these results indicate that the PDI57 peptide contains ER localization determinants recognized by a conserved machinery present in D. discoideum and Saccharomyces cerevisiae.

L8 ANSWER 14 OF 113 MEDLINE
AN 2000102705 MEDLINE
DN 20102705 PubMed ID: 10636893
TI Protein-disulfide isomerase (PDI) in FRTL5 cells. pH-dependent thyroglobulin/PDI interactions determine a novel PDI function in the post-endoplasmic reticulum of thyrocytes.
AU Mezghrani A; Courageot J; Mani J C; Pugniere M; Bastiani P; Miquelis R
CS Laboratoire de Biochimie, Ingenierie des Proteines, UMR 6560, Institut Federatif Jean Roche, Universite de la Mediterranee, Faculte de Medecine-Nord, Boulevard Pierre Dramard, 13916 Marseille Cedex 20, France.
SO JOURNAL OF BIOLOGICAL CHEMISTRY, (2000 Jan 21) 275 (3) 1920-9.
Journal code: HIV; 2985121R. ISSN: 0021-9258.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 200002
ED Entered STN: 20000309
Last Updated on STN: 20000309
Entered Medline: 20000224
AB Thyroglobulin (TG) is secreted by the thyrocytes into the follicular lumen of the thyroid. After maturation and hormone formation, TG is endocytosed and delivered to lysosomes. Quality control mechanisms may occur during this bidirectional traffic since 1) several molecular chaperones are cosecreted with TG in vivo, and 2) lysosomal targeting of immature TG is thought to be prevented via the interaction, in acidic conditions, between the Ser(789)-Met(1172) TG hormonogenic domain (BD) and an unidentified membrane receptor. We investigated the secretion and cell surface expression of PDI and other chaperones in the FRTL5 thyroid cell line, and then studied the characteristics of the interaction between TG and PDI. We demonstrated that PDI, but also other chaperones such as calnexin and ***KDEL*** -containing proteins are exposed at the cell surface. We observed on living cells or membrane preparations that PDI specifically binds TG in acidic conditions, and that only BD is involved in binding. Surface plasmon resonance analysis of TG/PDI interactions indicated: 1) that PDI bound TG but only in acidic conditions, and that it preferentially recognized immature molecules, and 2) BD is involved in binding even if cysteine-rich modules are deleted. The notion that PDI acts as an "escort" for immature TG in acidic post-endoplasmic reticulum compartments is discussed.

L8 ANSWER 15 OF 113 CAPLUS COPYRIGHT 2001 ACS
AN 2000:800591 CAPLUS
DN 134:70023
TI Production of hepatitis B surface antigen in transgenic plants for oral immunization
AU Richter, Liz J.; Thanavala, Yasmin; Arntzen, Charles J.; Mason, Hugh S.
CS Boyce Thompson Institutefor Plant Research, Inc, Ithaca, NY, 14853-1801, USA
SO Nat. Biotechnol. (2000), 18(11), 1167-1171
CODEN: NABIF9; ISSN: 1087-0156
PB Nature America Inc.
DT Journal
LA English
AB Here the authors present data showing oral immunogenicity of recombinant hepatitis B surface antigen (HBsAg) in preclin. animal trials. Mice fed transgenic HBsAg potato tubers showed a primary immune response (increases in HBsAg-specific serum antibody) that could be greatly boosted by i.p. delivery of a single subimmunogenic dose of com. HBsAg vaccine, indicating that plants expressing HBsAg in edible tissues may be a new means for oral hepatitis B immunization. However, attainment of such a goal will require higher HBsAg expression than was obsd. for the potatoes used in this study. The authors conducted a systematic anal. of factors influencing the accumulation of HBsAg in transgenic potato, including 5' and 3' flanking elements and protein targeting within plant cells. The most striking improvements resulted from (1) alternative polyadenylation signals, and (2) ***fusion*** proteins contg. targeting signals designed to enhance integration or retention of HBsAg in the endoplasmic reticulum (ER) of plant cells.
RE.CNT 30
RE
(2) An, G; Plant Cell 1989, V1, P115 CAPLUS
(4) Becker, D; Plant Mol Biol 1992, V20, P1195 CAPLUS
(5) Bednarek, S; Plant Mol Biol 1992, V20, P133 CAPLUS
(6) Bruss, V; Intervirology 1996, V39, P23 CAPLUS
(7) Chan, M; Proc Natl Acad Sci USA 1998, V95, P6543 CAPLUS
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L8 ANSWER 16 OF 113 CAPLUS COPYRIGHT 2001 ACS DUPLICATE 6
AN 2000:858290 CAPLUS
DN 135:163117
TI Accumulation of maize .gamma.-zein and .gamma.-zein: ***KDEL*** to high levels in tobacco leaves and differential increase of BiP synthesis in transformants
AU Bellucci, M.; Alpini, A.; Paolocci, F.; Cong, L.; Arcioni, S.
CS Istituto di Ricerche sul Miglioramento Genetico delle Piante Foraggere, CNR, Perugia, 06128, Italy
SO Theor. Appl. Genet. (2000), 101(5-6), 796-804
CODEN: THAGA6; ISSN: 0040-5752
PB Springer-Verlag
DT Journal
LA English
AB Two gene constructs (pROK.TG1L and pROK.TG1LK) were utilized to achieve accumulation of maize .gamma.-zein to high levels in tobacco (Nicotiana tabacum L.) leaves. Both the ***chimeric*** genes contained the .gamma.-zein-coding region preceded by the 5'-untranslated leader from the coat protein mRNA of TMV, but one of them (pROK.TG1LK) was modified in its protein-coding region by the addn. of the ER retention signal ***KDEL*** . The accumulation of .gamma.-zein and .gamma.-zein: ***KDEL*** in leaves was compared with ***heterologous*** protein accumulation in tobacco plants previously transformed with a .gamma.-zein cDNA harboring a native 5'UTR. Replacement of .gamma.-zein 5'UTR with the TMV leader dramatically increased .gamma.-zein prodn. Furthermore, .gamma.-zein: ***KDEL*** -expressing plants, on av., accumulated twice as much foreign protein in their leaves as pROK.TG1L plants. The two-fold increase in the level of .gamma.-zein: ***KDEL*** can probably be attributed to an improvement in the mechanism for ER retention of zeins in the transgenic cells. Transformants also showed increased prodn. of BiP, though to a lesser extent in .gamma.-zein: ***KDEL*** -expressing plants compared with pROK.TG1L plants. It is therefore likely that .gamma.-zein: ***KDEL*** retention is made less dependent on the chaperone assistance of BiP by the presence of the ***KDEL*** signal on the .gamma.-zein mutant.
RE.CNT 33
RE
(1) Bagga, S; Plant Cell 1997, V9, P1683 CAPLUS
(2) Bagga, S; Plant Physiol 1995, V107, P13 CAPLUS
(3) Barry, T; Br J Nutr 1981, V46, P521 CAPLUS
(4) Bellucci, M; Plant Sci 1997, V127, P161 CAPLUS
(5) Bellucci, M; Theor Appl Genet 1999, V98, P257 CAPLUS
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L8 ANSWER 17 OF 113 MEDLINE DUPLICATE 7
AN 2000229548 MEDLINE
DN 20229548 PubMed ID: 10764837
TI Two distinct domains of the beta-subunit of glucosidase II interact with the catalytic alpha-subunit.
AU Arendt C W; Ostergaard H L
CS Department of Medical Microbiology and Immunology, University of Alberta, Edmonton, Alberta T6G 2S2, Canada.
SO GLYCOBIOLOGY, (2000 May) 10 (5) 487-92.
Journal code: BEL; 9104124. ISSN: 0959-6658.
CY ENGLAND: United Kingdom
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 200005
ED Entered STN: 20000518
Last Updated on STN: 20000518
Entered Medline: 20000505
AB Recent purification and cDNA cloning of the endoplasmic reticulum processing enzyme glucosidase II have revealed that it is composed of two soluble proteins: a catalytic alpha-subunit and a beta-subunit of unknown function, both of which are highly conserved in mammals. Since the beta-subunit, which contains a C-terminal His-Asp-Glu-Leu (HDEL) motif, may function to link the catalytic subunit to the ***KDEL*** receptor as a retrieval mechanism, we sought to map the regions of the mouse beta-subunit protein responsible for mediating the association with the alpha-subunit. By screening a panel of recombinant beta-subunit glutathione S-transferase ***fusion*** proteins for the ability to precipitate glucosidase II activity, we have identified two non-overlapping interaction domains (ID1 and ID2) within the beta-subunit. ID1 encompasses 118 amino acids at the N-terminus of the mature polypeptide, spanning the cysteine-rich element in this region. ID2, located near the C-terminus, is contained within amino acids 273-400, a region occupied in part by a stretch of acidic residues. Variable usage of 7 alternatively spliced amino acids within ID2 was found not to influence the association of the two sub-units. We theorize that the catalytic subunit of glucosidase II binds synergistically to ID1 and ID2, explaining the high associative stability of the enzyme complex.

L8 ANSWER 18 OF 113 MEDLINE DUPLICATE 8
AN 2000193011 MEDLINE
DN 20193011 PubMed ID: 10730768

TI Development of ***chimeric*** molecules for recognition and targeting of antigen-specific B cells in pemphigus vulgaris.
CM Comment in: Br J Dermatol. 2000 Feb;142(2):208-9
AU Proby C M; Ota T; Suzuki H; Koyasu S; Gamou S; Shimizu N; Wahl J K; Wheelock M J; Nishikawa T; Amagai M
CS Department of Dermatology, Keio University School of Medicine, Tokyo, Japan.
SO BRITISH JOURNAL OF DERMATOLOGY, (2000 Feb) 142 (2) 321-30. Journal code: AW0; 0004041. ISSN: 0007-0963.
CY ENGLAND: United Kingdom
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 200004
ED Entered STN: 20000505
Last Updated on STN: 20000505
Entered Medline: 20000427
AB Pemphigus vulgaris (PV) is an autoimmune blistering disease characterized by circulating pathogenic IgG antibodies against desmoglein 3 (Dsg3). The purpose of this study was to develop ***chimeric*** molecules for specific recognition and elimination of autoimmune B cells in PV. Mouse hybridoma cell lines producing anti-Dsg3 antibody (5H10, 12A2) were developed as an in vitro model system for targeting B cells. Dsg3-GFP, a baculoprotein containing the entire extracellular domain of Dsg3 fused with green fluorescence protein, recognized and targeted the hybridoma cells through their surface immunoglobulin receptors in an antigen-specific way. The epitopes of these monoclonal antibodies were mapped on the amino terminal EC1 and part of EC2, a region considered functionally important in cadherins. ***Chimeric*** toxin molecules containing the mapped region (Dsg3deltaN1) and modified Pseudomonas exotoxin were produced in bacteria (Dsg3deltaN1-PE40- ***KDEL*** , PE3 7-Dsg3deltaN1- ***KDEL***) and tested in vitro on hybridoma cell lines. The ***chimeric*** toxins, but not Dsg3deltaN1 alone, showed dose-dependent toxic activity with a reduction in hybridoma cell number to 40-60% of toxin-negative control cultures, compared with little or no effect on anti-Dsg3-negative hybridoma cells. Furthermore, these toxins showed toxic effects on anti-Dsg3 IgG-producing B cells from Dsg3deltaN1-immunized mice, with a 60% reduction in cell number compared with Dsg3deltaN1 alone. Thus, specific recognition and targeting of antigen-specific B cells in PV was demonstrated; this strategy may hold promise as a future therapeutic option for PV and other autoimmune diseases.

L8 ANSWER 19 OF 113 CAPLUS COPYRIGHT 2001 ACS DUPLICATE 9
AN 2001:96365 CAPLUS
TI Expression of maize .gamma.-zein and .beta.-zein genes in transgenic Nicotiana tabacum and Lotus corniculatus
AU Bellucci, Michele; Alpini, Angelica; Arcioni, Sergio
CS Istituto di Ricerche sul Miglioramento Genetico delle Piante Foraggere (IRMGPF), CNR, Perugia, 06128, Italy
SO Plant Cell, Tissue Organ Cult. (2000), 62(2), 141-151
CODEN: PTCEDJ; ISSN: 0167-6857
PB Kluwer Academic Publishers
DT Journal
LA English
AB Accumulation of zeins, the endosperm storage proteins of maize, in a ***heterologous*** plant expression system was attempted. Plants of Nicotiana tabacum and Lotus corniculatus were transformed by Agrobacterium with binary vectors harbouring genes that code for .gamma.-zein and .beta.-zein, two zeins rich in sulfur amino acids. Adding the ER retention signal ***KDEL*** to the C-terminal domain modified the zein polypeptides. Significant levels of .gamma.-zein: ***KDEL*** and .beta.-zein: ***KDEL*** were detected in primary transformants of tobacco. Moreover, the two zeins colocalized in leaf protein bodies of .gamma.-l.beta.-zein: ***KDEL*** plants derived from a cross between two primary transformants. Coexpression of .gamma.-zein: ***KDEL*** and .beta.-zein: ***KDEL*** could be a useful strategy to obtain genotypes of forage legumes which are able to accumulate sulfur amino acids to high levels. As a first step, L. corniculatus plants expressing .gamma.-zein: ***KDEL*** in the leaves were obtained.
RE.CNT 29
RE
(1) Alvarez, I; Planta 1998, V205, P420 CAPLUS
(2) Bagga, S; Plant Cell 1997, V9, P1683 CAPLUS
(3) Bagga, S; Plant Physiol 1995, V107, P13 CAPLUS
(5) Bellucci, M; Plant Sci 1997, V127, P161 CAPLUS
(6) Bellucci, M; Theor Appl Genet 1999, V98, P257 CAPLUS
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L8 ANSWER 20 OF 113 CAPLUS COPYRIGHT 2001 ACS
AN 1999:753107 CAPLUS
DN 131:350254
TI Verotoxin B subunit for immunization
IN Green, Allan M.
PA USA
SO PCT Int. Appl., 47 pp.
CODEN: PIXXD2
DT Patent
LA English
FAN.CNT 1

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI WO 9959627	A2	19991125	WO 1999-US10679	19990514
WO 9959627	A3	20000120		
W: AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, SD, SL, SZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG				
AU 9939918	A1	19991206	AU 1999-39918	19990514
EP 1078007	A2	20010228	EP 1999-923063	19990514
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, FI				
PRAI US 1998-85693	P	19980515		
WO 1999-US10679	W	19990514		

AB The author discloses methods for stimulating an immune response in a mammal by administering a toxin-antigen conjugate. In one example, a ***fusion*** construct of a MAGE-1 epitope and the B subunit of verotoxin was shown to undergo processing and MHC class I presentation by APC and to stimulate cytotoxic T-cells.

L8 ANSWER 21 OF 113 CAPLUS COPYRIGHT 2001 ACS
AN 2000:559644 CAPLUS
DN 133:131182
TI Insecticidal ***fusion*** protein, its coded gene and method for producing transgenosis strain using said gene
IN Zhu, Zhen; Deng, Chaoyang; Qu, Qiang
PA Genetics Inst., Chinese Academy of Sciences, Peop. Rep. China
SO Faming Zhuanli Shenqing Gongkai Shuomingshu, 55 pp.
CODEN: CNXXEV
DT Patent
LA Chinese
FAN.CNT 1

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI CN 1229087	A	19990922	CN 1999-103430	19990330

AB The disclosed insecticidal ***fusion*** protein contains signal peptide at its N-terminal, insecticidal protein, and endoplasmic reticulum-retention signal at its C-terminal. The signal peptide is selected from potato patatin signal peptide, pathogenesis-related protein PR signal peptide, and soybean Kunitz type trypsin inhibitor (SKTI) signal peptide; the insecticidal protein is selected from Bacillus thuringiensis (Bt) toxoprotein, cowpea trypsin inhibitor (CpTI) insect-resistant protein, paddy mercapto- protease inhibitor (OC), or bivalent insecticidal protein comprising their ***fusion*** proteins; and the signal peptide of the insecticidal protein and endoplasmic reticulum-retention signal such as ***KDEL*** and HDEL. The expression vector is a plant-transfecting vector, contains one or more insecticidal gene expression box and/or other gene expression box, and the exogenous gene of the expression box is controlled under plant promoter. The plant promoter is selected from CaMV 35S promoter, CLCuV replicase gene promoter, paddy actin promoter, T-DNA mas promoter, maize ubiquitin promoter, and their promoter complexes. The expression vector is used for prepn. of insect-resistant plants such as paddy, maize, wheat, tobacco, cotton, soybean, potato, cabbage, brassica oleracea, and pepper, etc. The transgenosis plant is prepd. by construction of expression vector encoding insecticidal ***fusion*** protein, transfecting plant cells with the vector, and culturing the plant cells.

L8 ANSWER 22 OF 113 MEDLINE
AN 2000036595 MEDLINE
DN 20036595 PubMed ID: 10567425
TI Dependence of ricin toxicity on translocation of the toxin A-chain from the endoplasmic reticulum to the cytosol.
AU Wesche J; Rapak A; Olsnes S
CS Institute for Cancer Research, The Norwegian Radium Hospital, Montebello, 0310 Oslo, Norway.
SO JOURNAL OF BIOLOGICAL CHEMISTRY, (1999 Nov 26) 274 (48) 34443-9. Journal code: HIV; 2985121R. ISSN: 0021-9258.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 199912
ED Entered STN: 20000113
Last Updated on STN: 20000113
Entered Medline: 19991229
AB Ricin acts by translocating to the cytosol the enzymatically active toxin A-chain, which inactivates ribosomes. Retrograde intracellular transport and translocation of ricin was studied under conditions that alter the sensitivity of cells to the toxin. For this purpose tyrosine sulfation of mutant A-chain in the Golgi apparatus, glycosylation in the endoplasmic reticulum (ER) and appearance of A-chain in the cytosolic fraction was monitored. Introduction of an ER retrieval signal, a C-terminal ***KDEL*** sequence, into the A-chain increased the toxicity and resulted in more efficient glycosylation, indicating enhanced transport from Golgi to ER. Calcium depletion inhibited neither sulfation nor glycosylation but inhibited translocation and toxicity, suggesting that the toxin is translocated to the cytosol by the pathway used by misfolded proteins that are targeted to the proteasomes for degradation. Slightly acidified medium had a similar effect. The proteasome inhibitor, lactacystin, sensitized cells to ricin and increased the amount of ricin

A-chain in the cytosol. Anti-Sec61alpha precipitated sulfated and glycosylated ricin A-chain, suggesting that retrograde toxin translocation involves Sec61p. The data indicate that retrograde translocation across the ER membrane is required for intoxication.

L8 ANSWER 23 OF 113 MEDLINE DUPLICATE 10
AN 2000036555 MEDLINE
DN 20036555 PubMed ID: 10567385
TI Overexpression of cyclooxygenase-2 induces cell cycle arrest. Evidence for a prostaglandin-independent mechanism.
AU Trifan O C; Smith R M; Thompson B D; Hla T
CS Center for Vascular Biology, Department of Physiology, University of Connecticut Health Center, Farmington, Connecticut 06030-3505, USA.
NC HL49094 (NHLBI)
HL54710 (NHLBI)
SO JOURNAL OF BIOLOGICAL CHEMISTRY, (1999 Nov 26) 274 (48) 34141-7.
Journal code: HIV; 2985121R. ISSN: 0021-9258.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 199912
ED Entered STN: 20000113
Last Updated on STN: 20000113
Entered Medline: 19991229
AB The immediate-early gene cyclooxygenase 2 (Cox-2) is induced in a variety of hyperplastic pathological conditions, including rheumatoid arthritis and colorectal cancer. Although a causal role for Cox-2 has been proposed, mechanisms by which Cox-2 function contributes to the pathogenesis of hyperplastic disease are not well defined. We constructed a green fluorescent protein-tagged Cox-2 (Cox-2-GFP) to examine its effects on a variety of cell types upon overexpression. Subcellular localization and enzymatic and pharmacological properties of Cox-2-GFP polypeptide were indistinguishable from those of the wild-type Cox-2 polypeptide. Overexpression of the Cox-2-GFP or the Cox-2 polypeptide by transient transfection suppressed the population of cells in the S phase of the cell cycle, with a concomitant increase in G(0)/G(1) population. In contrast, transient overexpression of GFP had no effect on cell cycle distribution, whereas endoplasmic reticulum-retained GFP (GFP- ***KDEL***) overexpression was associated with only a minor decrease of cells in S phase. Interestingly, neither NS-398 (a Cox-2-specific inhibitor) nor indomethacin could reverse the effect of Cox-2-GFP overexpression on cell cycle progression. Furthermore, two mutants of Cox-2, S516Q and S516M, which lack the cyclooxygenase activity, exhibited the same effect as Cox-2-GFP. The cell cycle effect of Cox-2-GFP was observed in ECV-304, NIH 3T3, COS-7, bovine microvascular endothelial cells, and human embryonic kidney 293 cells. These findings suggest that Cox-2 inhibits cell cycle progression in a variety of cell types by a novel mechanism that does not require the synthesis of prostaglandins.

L8 ANSWER 24 OF 113 MEDLINE DUPLICATE 11
AN 1999386987 MEDLINE
DN 99386987 PubMed ID: 10455179
TI Molecular characterization of a novel basement membrane-associated proteoglycan, leprecan.
AU Wassenhove-McCarthy D J; McCarthy K J
CS Department of Pathology, School of Medicine, Louisiana State University Medical Center, Shreveport, Louisiana 71130, USA.
NC 1-RO1-DK48055 (NIDDK)
SO JOURNAL OF BIOLOGICAL CHEMISTRY, (1999 Aug 27) 274 (35) 25004-17.
Journal code: HIV; 2985121R. ISSN: 0021-9258.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
OS GENBANK-AF087433
EM 199909
ED Entered STN: 19991012
Last Updated on STN: 19991012
Entered Medline: 19990930
AB A monoclonal antibody was used in early studies to identify a novel chondroitin sulfate proteoglycan, secreted by L-2 cells, the core protein of which was approximately 100 kDa. To characterize this proteoglycan core protein at the molecular level, an L-2 cell cDNA library was probed by expression screening and solution hybridization. Northern blot analysis assigned transcript size to approximately 3.1 kilobases and, after contig assembly, the coding region of the mRNA corresponded to 2.18 kilobases. Immunoassays were performed to confirm the identity of this sequence, using a polyclonal antibody raised against an expressed ***fusion*** protein encoded by sequence representing the carboxyl half of the molecule. The antibody recognized the core protein in Western blots after prior digestion of the intact proteoglycan with chondroitinase ABC. Immunostaining tissue sections with the same antibody localized the proteoglycan to basement membranes, and expression of the entire sequence in Chinese hamster ovary K-1 cells showed that the protein encoded by the sequence secreted as a chondroitin sulfate proteoglycan. The core protein not only has motifs permitting glycosylation as a proteoglycan, but also possesses the endoplasmic reticulum retrieval signal, ***KDEL*** , which suggests that, in addition to its role as a basement membrane component, it may also participate in the secretory pathway of cells.

L8 ANSWER 25 OF 113 MEDLINE DUPLICATE 12
AN 1999134317 MEDLINE
DN 99134317 PubMed ID: 9933586
TI Structural basis for the differential toxicity of cholera toxin and Escherichia coli heat-labile enterotoxin. Construction of ***hybrid*** toxins identifies the A2-domain as the determinant of differential toxicity.
AU Rodighiero C; Aman A T; Kenny M J; Moss J; Lencer W I; Hirst T R
CS Department of Pathology and Microbiology, University of Bristol, School of Medical Sciences, Bristol BS8 1TD, United Kingdom.
SO JOURNAL OF BIOLOGICAL CHEMISTRY, (1999 Feb 12) 274 (7) 3962-9.
Journal code: HIV; 2985121R. ISSN: 0021-9258.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 199903
ED Entered STN: 19990324
Last Updated on STN: 19990324
Entered Medline: 19990311
AB Cholera toxin (Ctx) and E. coli heat-labile enterotoxin (Etx) are structurally and functionally similar AB5 toxins with over 80% sequence identity. When their action in polarized human epithelial (T84) cells was monitored by measuring toxin-induced Cl- ion secretion, Ctx was found to be the more potent of the two toxins. Here, we examine the structural basis for this difference in toxicity by engineering a set of mutant and ***hybrid*** toxins and testing their activity in T84 cells. This revealed that the differential toxicity of Ctx and Etx was (i) not due to differences in the A-subunit's C-terminal ***KDEL*** targeting motif (which is RDEL in Etx), as a ***KDEL*** to RDEL substitution had no effect on cholera toxin activity; (ii) not attributable to the enzymatically active A1-fragment, as ***hybrid*** toxins in which the A1-fragment in Ctx was substituted for that of Etx (and vice versa) did not alter relative toxicity; and (iii) not due to the B-subunit, as the replacement of the B-subunit in Ctx for that of Etx caused no alteration in toxicity, thus excluding the possibility that the broader receptor specificity of EtxB is responsible for reduced activity. Remarkably, the difference in toxicity could be mapped to a 10-amino acid segment of the A2-fragment that penetrates the central pore of the B-subunit pentamer. A comparison of the in vitro stability of two ***hybrid*** toxins, differing only in this 10-amino acid segment, revealed that the Ctx A2-segment conferred a greater stability to the interaction between the A- and B-subunits than the corresponding segment from Etx A2. This suggests that the reason for the relative potency of Ctx compared with Etx stems from the increased ability of the A2-fragment of Ctx to maintain holotoxin stability during uptake and transport into intestinal epithelia.

L8 ANSWER 26 OF 113 MEDLINE
AN 1999249887 MEDLINE
DN 99249887 PubMed ID: 10233100
TI Diffusion of green fluorescent protein in the aqueous-phase lumen of endoplasmic reticulum.
AU Dayel M J; Hom E F; Verkman A S
CS Departments of Medicine and Physiology, Cardiovascular Research Institute, San Francisco, California 94143-0521, USA.
NC DK16095 (NIDDK)
DK43840 (NIDDK)
HL60288 (NHLBI)
+
SO BIOPHYSICAL JOURNAL, (1999 May) 76 (5) 2843-51.
Journal code: A5S; 0370626. ISSN: 0006-3495.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 199906
ED Entered STN: 19990712
Last Updated on STN: 19990712
Entered Medline: 19990621
AB The endoplasmic reticulum (ER) is the major compartment for the processing and quality control of newly synthesized proteins. Green fluorescent protein (GFP) was used as a noninvasive probe to determine the viscous properties of the aqueous lumen of the ER. GFP was targeted to the ER lumen of CHO cells by transient transfection with cDNA encoding GFP (S65T/F64L mutant) with a C-terminus ***KDEL*** retention sequence and upstream prolactin secretory sequence. Repeated laser illumination of a fixed 2-micrometers diameter spot resulted in complete bleaching of ER-associated GFP throughout the cell, indicating a continuous ER lumen. A residual amount (<1%) of GFP- ***KDEL*** was perinuclear and noncontiguous with the ER, presumably within a pre- or cis-Golgi compartment involved in ***KDEL*** -substrate retention. Quantitative spot photobleaching with a single brief bleach pulse indicated that GFP was fully mobile with a t1/2 for fluorescence recovery of 88 +/- 5 ms (SE; 60x lens) and 143 +/- 8 ms (40x). Fluorescence recovery was abolished by paraformaldehyde except for a small component of reversible photobleaching with t1/2 of 3 ms. For comparison, the t1/2 for photobleaching of GFP in cytoplasm was 14 +/- 2 ms (60x) and 24 +/- 1 ms (40x). Utilizing a mathematical model that accounted for ER reticular geometry, a GFP diffusion coefficient of 0.5-1 x 10(-7) cm2/s was computed, 9-18-fold less than that in water and 3-6-fold less than that in cytoplasm. By frequency-domain microfluorimetry, the GFP rotational correlation time was measured to be 39 +/- 8 ns, approximately 2-fold greater than that in water but comparable to that in the cytoplasm. Fluorescence recovery after photobleaching using a 40x lens was measured (at 23 degrees C unless otherwise indicated) for several potential effectors of ER structure

and/or lumen environment: t1/2 values (in ms) were 143 +/- 8 (control), 100 +/- 13 (37 degrees C), 53 +/- 13 (brefeldin A), and 139 +/- 6 (dithiothreitol). These results indicate moderately slowed GFP diffusion in a continuous ER lumen.

L8 ANSWER 27 OF 113 MEDLINE DUPLICATE 13
AN 1999173956 MEDLINE
DN 99173956 PubMed ID: 10074109
TI The transmembrane domain of hepatitis C virus glycoprotein E1 is a signal for static retention in the endoplasmic reticulum.
AU Cocquerel L; Duvet S; Meunier J C; Pillez A; Cacan R; Wychowski C; Dubuisson J
CS CNRS-UMR319, IBL/Institut Pasteur de Lille, 59021 Lille Cedex, France.
SO JOURNAL OF VIROLOGY, (1999 Apr) 73 (4) 2641-9.
Journal code: KCV; 0113724. ISSN: 0022-538X.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 199905
ED Entered STN: 19990517
Last Updated on STN: 19990517
Entered Medline: 19990506
AB Hepatitis C virus (HCV) glycoproteins E1 and E2 assemble to form a noncovalent heterodimer which, in the cell, accumulates in the endoplasmic reticulum (ER). Contrary to what is observed for proteins with a ***KDEL*** or a KKXX ER-targeting signal, the ER localization of the HCV glycoprotein complex is due to a static retention in this compartment rather than to its retrieval from the cis-Golgi region. A static retention in the ER is also observed when E2 is expressed in the absence of E1 or for a ***chimeric*** protein containing the ectodomain of CD4 in ***fusion*** with the transmembrane domain (TMD) of E2. Although they do not exclude the presence of an intracellular localization signal in E1, these data do suggest that the TMD of E2 is an ER retention signal for HCV glycoprotein complex. In this study ***chimeric*** proteins containing the ectodomain of CD4 or CD8 fused to the C-terminal hydrophobic sequence of E1 were shown to be localized in the ER, indicating that the TMD of E1 is also a signal for ER localization. In addition, these ***chimeric*** proteins were not processed by Golgi enzymes, indicating that the TMD of E1 is responsible for true retention in the ER, without recycling through the Golgi apparatus. Together, these data suggest that at least two signals (TMDs of E1 and E2) are involved in ER retention of the HCV glycoprotein complex.

L8 ANSWER 28 OF 113 MEDLINE DUPLICATE 14
AN 2000029763 MEDLINE
DN 20029763 PubMed ID: 10562278
TI Rab6 coordinates a novel Golgi to ER retrograde transport pathway in live cells.
CM Erratum in: J Cell Biol 2000 Jan 10;148(1):followi
AU White J; Johannes L; Mallard F; Girod A; Grill S; Reinsch S; Keller P; Tzschaschel B; Echard A; Goud B; Stelzer E H
CS Light Microscopy Group, European Molecular Biology Laboratory, Meyerhofstrasse 1, D-69117 Heidelberg, Germany.. jwhite@embl-heidelberg.de
SO JOURNAL OF CELL BIOLOGY, (1999 Nov 15) 147 (4) 743-60.
Journal code: HMV; 0375356. ISSN: 0021-9525.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 199912
ED Entered STN: 20000113
Last Updated on STN: 20000330
Entered Medline: 19991217
AB We visualized a fluorescent-protein (FP) ***fusion*** to Rab6, a Golgi-associated GTPase, in conjunction with fluorescent secretory pathway markers. FP-Rab6 defined highly dynamic transport carriers (TCs) translocating from the Golgi to the cell periphery. FP-Rab6 TCs specifically accumulated a retrograde cargo, the wild-type Shiga toxin B-fragment (STB), during STB transport from the Golgi to the endoplasmic reticulum (ER). FP-Rab6 TCs associated intimately with the ER, and STB entered the ER via specialized peripheral regions that accumulated FP-Rab6. Microinjection of antibodies that block coatomer protein I (COPI) function inhibited trafficking of a ***KDEL*** -receptor FP-***fusion*** , but not FP-Rab6. Additionally, markers of COPI-dependent recycling were excluded from FP-Rab6/STB TCs. Overexpression of Rab6:GDP (T27N mutant) using T7 vaccinia inhibited toxicity of Shiga holotoxin, but did not alter STB transport to the Golgi or Golgi morphology. Taken together, our results indicate Rab6 regulates a novel Golgi to ER transport pathway.

L8 ANSWER 29 OF 113 MEDLINE DUPLICATE 15
AN 2000030157 MEDLINE
DN 20030157 PubMed ID: 10561693
TI Calreticulin is transported to the surface of NG108-15 cells where it forms surface patches and is partially degraded in an acidic compartment.
AU Xiao G; Chung T F; Fine R E; Johnson R J
CS Department of Chemistry, Boston University, Boston, Massachusetts, USA.
NC R37 AG05894 (NIA)
SO JOURNAL OF NEUROSCIENCE RESEARCH, (1999 Dec 1) 58 (5) 652-62.
Journal code: KAC; 7600111. ISSN: 0360-4012.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 200001
ED Entered STN: 20000114
Last Updated on STN: 20000114
Entered Medline: 20000105
AB Although calreticulin (Crt) is primarily localized to the endoplasmic reticulum (ER), our results using biotinylation and immunocytochemical methods indicate that a small but significant amount of Crt is present and forms large patches on the surface of NG108-15 cells (a mouse neuroblastoma-rat glioma ***hybrid*** cell line). (35)S-labelled Crt molecules begin to reach the cell surface after only 10 min of labelling and disappear slowly from the cell surface. After 4 hr of labelling, approximately half of the newly synthesized Crt molecules are on the cell surface. We believe that some Crt molecules may escape from the ***KDEL*** receptor-mediated salvage pathway as they are synthesized and proceed through the secretory pathway to the cell surface. Immunoprecipitation from the culture medium shows that Crt is not released from the cell surface to the medium, suggesting tight binding to surface molecules. NH(4)Cl can block the degradation of Crt; therefore, Crt is presumably degraded in the lysosome pathway. However, blockage of the disappearance of surface Crt is less efficient than that of internal Crt. This suggests that the disappearance of Crt from the cell surface may not be due solely to its degradation, but may reflect transport into another cell compartment such as the ER.

L8 ANSWER 30 OF 113 MEDLINE
AN 1999115680 MEDLINE
DN 99115680 PubMed ID: 9914159
TI The ***KDEL*** retrieval system is exploited by Pseudomonas exotoxin A, but not by Shiga-like toxin-1, during retrograde transport from the Golgi complex to the endoplasmic reticulum.
AU Jackson M E; Simpson J C; Girod A; Pepperkok R; Roberts L M; Lord J M
CS Department of Biological Sciences, University of Warwick, Coventry, CV4 7AL, UK.. ml@dna.bio.warwick.ac.uk
SO JOURNAL OF CELL SCIENCE, (1999 Feb) 112 (Pt 4) 467-75.
Journal code: HNK; 0052457. ISSN: 0021-9533.
CY ENGLAND: United Kingdom
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 199907
ED Entered STN: 19990730
Last Updated on STN: 19990730
Entered Medline: 19990720
AB To investigate the role of the ***KDEL*** receptor in the retrieval of protein toxins to the mammalian cell endoplasmic reticulum (ER), lysozyme variants containing AARL or ***KDEL*** C-terminal tags, or the human ***KDEL*** receptor, have been expressed in toxin-treated COS 7 and HeLa cells. Expression of the lysozyme variants and the ***KDEL*** receptor was confirmed by immunofluorescence. When such cells were challenged with diphtheria toxin (DT) or Escherichia coli Shiga-like toxin 1 (SLT-1), there was no observable difference in their sensitivities as compared to cells which did not express these exogenous proteins. By contrast, the cytotoxicity of Pseudomonas exotoxin A (PE) is reduced by expressing lysozyme- ***KDEL*** , which causes a redistribution of the ***KDEL*** receptor from the Golgi complex to the ER, and cells are sensitised to this toxin when they express additional ***KDEL*** receptors. These data suggest that, in contrast to SLT-1, PE can exploit the ***KDEL*** receptor in order to reach the ER lumen where it is believed that membrane transfer to the cytosol occurs. This contention was confirmed by microinjecting into Vero cells antibodies raised against the cytoplasmically exposed tail of the ***KDEL*** receptor. Immunofluorescence confirmed that these antibodies prevented the retrograde transport of the ***KDEL*** receptor from the Golgi complex to the ER, and this in turn reduced the cytotoxicity of PE, but not that of SLT-1, to these cells.

L8 ANSWER 31 OF 113 MEDLINE DUPLICATE 16
AN 1999135889 MEDLINE
DN 99135889 PubMed ID: 9950687
TI Morphological and functional association of Sec22b/ERS-24 with the pre-Golgi intermediate compartment.
AU Zhang T; Wong S H; Tang B L; Xu Y; Hong W
CS Membrane Biology Laboratory, Institute of Molecular and Cell Biology, Singapore 117609, Singapore.
SO MOLECULAR BIOLOGY OF THE CELL, (1999 Feb) 10 (2) 435-53.
Journal code: BAU; 9201390. ISSN: 1059-1524.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 199903
ED Entered STN: 19990326
Last Updated on STN: 19990326
Entered Medline: 19990318
AB Yeast Sec22p participates in both anterograde and retrograde vesicular transport between the endoplasmic reticulum (ER) and the Golgi apparatus by functioning as a v-SNARE (soluble N-ethylmaleimide-sensitive factor [NSF] attachment protein receptor) of transport vesicles. Three mammalian proteins homologous to Sec22p have been identified and are referred to as Sec22a, Sec22b/ERS-24, and Sec22c, respectively. The existence of three

homologous proteins in mammalian cells calls for detailed cell biological and functional examinations of each individual protein. The epitope-tagged forms of all three proteins have been shown to be primarily associated with the ER, although functional examination has not been carefully performed for any one of them. In this study, using antibodies specific for Sec22b/ERS-24, it is revealed that endogenous Sec22b/ERS-24 is associated with vesicular structures in both the perinuclear Golgi and peripheral regions. Colabeling experiments for Sec22b/ERS-24 with Golgi mannosidase II, the ***KDEL*** receptor, and the envelope glycoprotein G (VSVG) of vesicular stomatitis virus (VSV) en route from the ER to the Golgi under normal, brefeldin A, or nocodazole-treated cells suggest that Sec22b/ERS-24 is enriched in the pre-Golgi intermediate compartment (IC). In a well-established semi-intact cell system that reconstitutes transport from the ER to the Golgi, transport of VSVG is inhibited by antibodies against Sec22b/ERS-24. EGTA is known to inhibit ER-Golgi transport at a stage after vesicle/transport intermediate docking but before the actual ***fusion*** event. Antibodies against Sec22b/ERS-24 inhibit ER-Golgi transport only when they are added before the EGTA-sensitive stage. Transport of VSVG accumulated in pre-Golgi IC by incubation at 15 degreesC is also inhibited by Sec22b/ERS-24 antibodies. Morphologically, VSVG is transported from the ER to the Golgi apparatus via vesicular intermediates that scatter in the peripheral as well as the Golgi regions. In the presence of antibodies against Sec22b/ERS-24, VSVG is seen to accumulate in these intermediates, suggesting that Sec22b/ERS-24 functions at the level of the IC in ER-Golgi transport.

L8 ANSWER 32 OF 113 MEDLINE
AN 2000115640 MEDLINE
DN 20115640 PubMed ID: 10648938
TI Inhibition of expression of the Galalpha1-3Gal epitope on porcine cells using an intracellular single-chain antibody directed against alpha1,3galactosyltransferase.
AU Sepp A; Farrar C A; Dorling T; Cairns T; George A J; Lechler R I
CS Department of Immunology, Division of Medicine, Imperial College School of Medicine, Hammersmith Campus, Du Cane Road, London, UK.
SO JOURNAL OF IMMUNOLOGICAL METHODS, (1999 Dec 10) 231 (1-2) 191-205.
Journal code: IFE; 1305440. ISSN: 0022-1759.
CY Netherlands
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 200002
ED Entered STN: 20000314
Last Updated on STN: 20000314
Entered Medline: 20000228
AB The carbohydrate epitope Galalpha1-3Gal has been shown to be the major target of natural antibodies responsible for hyperacute rejection of porcine tissues transplanted into primates. We have sought to produce a phenotypic knockout of the alpha1, 3Galactosyltransferase enzyme that is responsible for generating this epitope, using an intracellular antibody approach. We have isolated high affinity anti-alpha1,3Galactosyltransferase single-chain antibodies from a semi-synthetic phage display library. Expression of a ***KDEL*** -tagged anti-alpha1,3Galactosyltransferase single-chain antibody in a porcine endothelial cell line resulted in the decreased expression of the Galalpha1-3Gal epitope and increased resistance to lysis by human serum.

L8 ANSWER 33 OF 113 MEDLINE DUPLICATE 17
AN 1999131391 MEDLINE
DN 99131391 PubMed ID: 9934692
TI Alternative mechanisms of interaction between homotypic and heterotypic parainfluenza virus HN and F proteins.
AU Tong S; Compans R W
CS Department of Microbiology and Immunology, Emory University, Atlanta, GA 30322, USA.
NC CA 18611 (NCI)
SO JOURNAL OF GENERAL VIROLOGY, (1999 Jan) 80 (Pt 1) 107-15.
Journal code: I9B; 0077340. ISSN: 0022-1317.
CY ENGLAND: United Kingdom
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 199903
ED Entered STN: 19990324
Last Updated on STN: 19990324
Entered Medline: 19990310
AB Cell ***fusion*** by human parainfluenza virus (HPIV) type 2 or type 3 requires the coexpression of both the ***fusion*** (F) and haemagglutinin-neuraminidase (HN) glycoproteins from the same virus type, indicating that promotion of ***fusion*** requires a type-specific interaction between F and HN. In this report we have further investigated the interaction of the ectodomains of the F and HN glycoproteins from HPIV2 and HPIV3. We constructed mutants of the HPIV2 F and HPIV3 F proteins (F'- ***KDEL***) lacking a transmembrane anchor and a cytoplasmic tail, and containing a C-terminal signal for retention in the endoplasmic reticulum (ER). The P12 and P13 F'- ***KDEL*** proteins were both found to be retained intracellularly, and neither could induce cell ***fusion*** when co-expressed with homotypic HN proteins. Qualitative and quantitative cell- ***fusion*** assays also showed that both the P12 F'- ***KDEL*** and P13 F'- ***KDEL*** proteins have inhibitory effects on P12 F- and HN-induced cell ***fusion*** . However, the F- ***KDEL*** mutants were found to inhibit cell ***fusion*** by two distinct mechanisms. An interaction between P12 F'- ***KDEL*** and P12 HN results in intracellular retention of HN, and a block in its transport to the cell surface. In contrast, P13 F'- ***KDEL*** was found to suppress the steady-state intracellular expression levels of HPIV2 HN. These results support the conclusion that ***fusion*** involves an interaction between the HN and F proteins, and suggest that an association between F and HN may occur in the ER.

L8 ANSWER 34 OF 113 CAPLUS COPYRIGHT 2001 ACS DUPLICATE 18
AN 1999:691352 CAPLUS
DN 132:76053
TI Accumulation of antibody ***fusion*** proteins in the cytoplasm and ER of plant cells
AU Spiegel, H.; Schillberg, S.; Sack, M.; Holzem, A.; Nahring, J.; Monecke, M.; Liao, Y.-C.; Fischer, R.
CS Institut fur Biologie I (Botanik/Molekulargenetik), RWTH Aachen, Aachen, D-52074, Germany
SO Plant Sci. (Shannon, Irel.) (1999), 149(1), 63-71
CODEN: PLSCE4; ISSN: 0168-9452
PB Elsevier Science Ireland Ltd.
DT Journal
LA English
AB To test whether the accumulation of cytoplasmically targeted recombinant antibodies could be improved by ***fusion*** to a cytoplasmic protein, we generated a series of single chain antibody- ***fusion*** proteins and assayed the levels of functional protein. Glutathione S-transferase (GST) from Schistosoma japonicum, coat protein (CP) from TMV, thioredoxin from tobacco (TRXt) or thioredoxin from Escherichia coli (TRXe) was fused to the N-terminus of scFv24, a TMV specific single chain antibody. Accumulation of functional ***fusion*** proteins in the endoplasmic reticulum (ER) and plant cell cytoplasm was analyzed by transient expression in tobacco leaves. ELISA anal. demonstrated that the ***fusion*** partners did not prevent the binding of scFv24 to TMV virions. However, accumulation of functional scFv24 was dependent on the ***fusion*** partner coupled to it. CP-scFv and GST-scFv ***fusion*** protein accumulation amounted to 1 .mu.g and 3 .mu.g/g of leaf material, resp., whereas the thioredoxin ***fusion*** proteins were produced at low levels. Western blot and surface plasmon resonance anal. confirmed the integrity of the ER retained CP and GST ***fusion*** proteins. In the cytoplasm, only the CP ***fusion*** protein was detectable (1-5 ng/g of leaf material) and levels of scFv24 alone or fused to the other three ***fusion*** partners were below the ELISA detection limit. Addn. of a ***KDEL*** sequence to the C-terminus of the cytoplasmic CP ***fusion*** resulted in a 3-fold increase in protein accumulation indicating that an N-terminal CP and the C-terminal ***KDEL*** sequence are suitable elements to stabilize scFv antibodies in the cytoplasm.
RE.CNT 40
RE
(1) Artsaenko, O; Plant J 1995, V8, P745 CAPLUS
(2) Asakura, I; Gastroenterol Jpn 1993, V28, P34 CAPLUS
(3) Biocca, S; Biotechnology 1995, V13, P1110 CAPLUS
(5) Brugidou, C; Mol Gen Genet 1993, V238, P285 CAPLUS
(6) Conrad, U; Plant Mol Biol 1998, V38, P101 CAPLUS
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L8 ANSWER 35 OF 113 CAPLUS COPYRIGHT 2001 ACS
AN 1998:568850 CAPLUS
DN 129:185085
TI Modified prodomain C-terminus of human carboxypeptidase B that enhances recombinant expression of the mature enzyme
IN Edge, Michael Derek
PA Zeneca Limited, UK
SO PCT Int. Appl., 88 pp.
CODEN: PIXXD2
DT Patent
LA English
FAN.CNT 1

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI WO 9835988	A1	19980820	WO 1998-GB415	19980210
W: AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GE, GH, GM, GW, HU, ID, IL, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, US, UZ, VN, YU, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, SD, SZ, UG, ZW, AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG				
AU 9860006	A1	19980908	AU 1998-60006	19980210
PRAI GB 1997-3104		19970214		
GB 1997-22003		19971018		
GB 1997-22727		19971029		
WO 1998-GB415		19980210		

AB The field of the invention is recombinant prodn. of carboxypeptidase B. This invention provides a modified prodomain of carboxypeptidase B which enhances recombinant expression thereof when co-expressed from a sep. gene. Preferred modified prodomains (residues 1-95 of the proenzyme) have added amino acids at their C-terminus, in particular any one of the following sequences: L, ***KDEL*** , KKAA or SDYQRL. The carboxypeptidase is preferably human pancreatic carboxypeptidase B. The invention also relates to corresponding polynucleotide sequences, vectors, host cells and methods of recombinant carboxypeptidase B prodn.

Expression of mature human pancreatic carboxypeptidase B from COS cells is enhanced by co-secretion of the modified prodomain. An esp. preferred carboxypeptidase B ***fusion*** construct comprises a gene encoding a humanized Fd heavy chain fragment of antibody 806.077 linked to [A248S,G251T,D253K]-human carboxypeptidase B and its co-expression with a gene encoding a humanized light chain of 806.077 and a gene encoding the pro-Leu modified prodomain of human carboxypeptidase B to give the F(ab')2 protein with a mol. of [A248S,G251T,D253K]carboxypeptidase B at the C-terminus of each of the heavy chain fragments. The const. and hinge regions of the humanized Fd heavy chain fragment are derived from the human IgG3 antibody type.

L8 ANSWER 36 OF 113 MEDLINE DUPLICATE 19
AN 1998337780 MEDLINE
DN 98337780 PubMed ID: 9671507
TI Interaction between a Ca2+-binding protein calreticulin and perforin, a component of the cytotoxic T-cell granules.
AU Andrin C; Pinkoski M J; Burns K; Atkinson E A; Krahenbuhl O; Hudig D; Fraser S A; Winkler U; Tschopp J; Opas M; Bleackley R C; Michalak M
CS Molecular Biology of Membranes Research Group, University of Alberta, Edmonton, Canada.
NC R01 CA38942 (NCI)
T32 CA09563 (NCI)
SO BIOCHEMISTRY, (1998 Jul 21) 37 (29) 10386-94.
Journal code: A0G; 0370623. ISSN: 0006-2960.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 199808
ED Entered STN: 19980817
Last Updated on STN: 19980817
Entered Medline: 19980805
AB Calreticulin is a component of cytotoxic T-lymphocyte and NK lymphocyte granules. We report here that granule-associated calreticulin terminates with the ***KDEL*** endoplasmic reticulum retrieval amino acid sequence and somehow escapes the ***KDEL*** retrieval system. In perforin knock-out mice calreticulin is still targeted into the granules. Thus, calreticulin will traffic without perforin to cytotoxic granules. In the granules, calreticulin and perforin are associated as documented by (i) copurification of calreticulin with perforin but not with granzymes and (ii) immunoprecipitation of a calreticulin-perforin complex using specific antibodies. By using calreticulin affinity chromatography and protein ligand blotting we show that perforin binds to calreticulin in the absence of Ca2+ and the two proteins dissociate upon exposure to 0.1 mM or higher Ca2+ concentration. Perforin interacts strongly with the P-domain of calreticulin (the domain which has high Ca2+-binding affinity and chaperone function) as revealed by direct protein-protein interaction, ligand blotting, and the yeast two- ***hybrid*** techniques. Our results suggest that calreticulin may act as Ca2+-regulated chaperone for perforin. This action will serve to protect the CTL during biogenesis of granules and may also serve to regulate perforin lytic action after release.

L8 ANSWER 37 OF 113 MEDLINE DUPLICATE 20
AN 1998363224 MEDLINE
DN 98363224 PubMed ID: 9699644
TI Modulation of apoptotic response of a radiation-resistant human carcinoma by Pseudomonas exotoxin- ***chimeric*** protein.
AU Seetharam S; Nodzenski E; Beckett M A; Heimann R; Cha A; Margulies I; Pastan I; Kufe D W; Weichselbaum R R
CS Department of Radiation and Cellular Oncology, University of Chicago Hospitals, Illinois 60637, USA.
NC CA-42596 (NCI)
CA-55241 (NCI)
SO CANCER RESEARCH, (1998 Aug 1) 58 (15) 3215-20.
Journal code: CNF; 2984705R. ISSN: 0008-5472.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 199808
ED Entered STN: 19980903
Last Updated on STN: 20000303
Entered Medline: 19980827
AB Strategies to sensitize human tumors that are resistant to apoptosis have been clinically unsuccessful. We demonstrate that a structurally modified ***chimeric*** Pseudomonas exotoxin, PEdelta53L/TGF-alpha/ ***KDEL*** , with binding specificity for the epidermal growth factor receptor, markedly enhances sensitivity of human xenografts to radiation killing. Exposure to PEdelta53L/TGF-alpha/ ***KDEL*** decreases the apoptotic threshold through protein synthesis inhibition and simultaneous production of ceramide in tumor cells that lack functional p53 protein. In contrast, no increase in local or systemic toxicity was observed with the ***chimeric*** toxin and radiation. We conclude that biochemical targeting of the ***chimeric*** toxin and physical targeting of ionizing radiation may increase the therapeutic ratio in the treatment of human cancers with alterations of p53 expression. This strategy offers a high therapeutic potential for Pseudomonas exotoxin A ***chimeric*** proteins and irradiation.

L8 ANSWER 38 OF 113 MEDLINE DUPLICATE 21
AN 1998425531 MEDLINE
DN 98425531 PubMed ID: 9754560
TI Major histocompatibility complex class I presentation of exogenous soluble tumor antigen fused to the B-fragment of Shiga toxin.
AU Lee R S; Tartour E; van der Bruggen P; Vantomme V; Joyeux I; Goud B; Fridman W H; Johannes L
CS Laboratoire d'Immunologie Clinique, INSERM U255, Institut Curie, Paris, France.
SO EUROPEAN JOURNAL OF IMMUNOLOGY, (1998 Sep) 28 (9) 2726-37.
Journal code: EN5; 1273201. ISSN: 0014-2980.
CY GERMANY: Germany, Federal Republic of
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 199810
ED Entered STN: 19981021
Last Updated on STN: 19981021
Entered Medline: 19981013
AB Targeting exogenous antigen into the MHC class I-restricted presentation pathway is a prerequisite for the induction of cytotoxic T lymphocytes (CTL) which have been shown to represent an important component of the protective and therapeutic immune response to viral infections and tumors. In this study, we produced recombinant proteins composed of the receptor-binding non-toxic B-fragment of bacterial Shiga toxin derived from Shigella dysenteriae associated with an epitope from a model tumor antigen, Mage 1. We show that Shiga B-Mage 1 ***fusion*** proteins carrying an active or inactive endoplasmic reticulum retrieval signal (the C-terminal peptides ***KDEL*** or KDELGL, respectively) could be presented by peripheral blood mononuclear cells in an MHC class I-restricted manner to Mage 1-specific CTL. After pulsing B lymphoblastoid cells or dendritic cells with Shiga B-Mage 1 ***fusion*** protein, activation of the MHC class I-restricted Mage 1-specific CTL was also demonstrated. In further analysis, we showed that treatment with brefeldin A or paraformaldehyde fixation of Epstein-Barr virus-transformed B cells prevented the presentation of the Mage 1 T cell epitope, which excluded extracellular processing of the antigen. Immunofluorescence analysis also revealed that the Shiga B-Mage 1 ***fusion*** protein was largely excluded from Lamp-2-positive lysosomal structures. Therefore, the ability of Shiga toxin B-fragment to target dendritic cells and B cells and to direct antigen into the exogenous class I-restricted pathway makes it an attractive non-living and non-toxic vaccine vector.

L8 ANSWER 39 OF 113 MEDLINE
AN 1999069482 MEDLINE
DN 99069482 PubMed ID: 9852151
TI Role of xklp3, a subunit of the Xenopus kinesin II heterotrimeric complex, in membrane transport between the endoplasmic reticulum and the Golgi apparatus.
AU Le Bot N; Antony C; White J; Karsenti E; Vernos I
CS Cell Biology and Biophysics Program, European Molecular Biological Laboratory, D-69117 Heidelberg, Germany.
SO JOURNAL OF CELL BIOLOGY, (1998 Dec 14) 143 (6) 1559-73.
Journal code: HMV; 0375356. ISSN: 0021-9525.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 199901
ED Entered STN: 19990209
Last Updated on STN: 19990209
Entered Medline: 19990126
AB The function of the Golgi apparatus is to modify proteins and lipids synthesized in the ER and sort them to their final destination. The steady-state size and function of the Golgi apparatus is maintained through the recycling of some components back to the ER. Several lines of evidence indicate that the spatial segregation between the ER and the Golgi apparatus as well as trafficking between these two compartments require both microtubules and motors. We have cloned and characterized a new Xenopus kinesin like protein, Xklp3, a subunit of the heterotrimeric Kinesin II. By immunofluorescence it is found in the Golgi region. A more detailed analysis by EM shows that it is associated with a subset of membranes that contain the ***KDEL*** receptor and are localized between the ER and Golgi apparatus. An association of Xklp3 with the recycling compartment is further supported by a biochemical analysis and the behavior of Xklp3 in BFA-treated cells. The function of Xklp3 was analyzed by transfecting cells with a dominant-negative form lacking the motor domain. In these cells, the normal delivery of newly synthesized proteins to the Golgi apparatus is blocked. Taken together, these results indicate that Xklp3 is involved in the transport of tubular-vesicular elements between the ER and the Golgi apparatus.

L8 ANSWER 40 OF 113 MEDLINE DUPLICATE 22
AN 1998401141 MEDLINE
DN 98401141 PubMed ID: 9731188
TI Differences in cytotoxicity of native and engineered RIPs can be used to assess their ability to reach the cytoplasm.
AU Svinth M; Steighardt J; Hernandez R; Suh J K; Kelly C; Day P; Lord M; Girbes T; Robertus J D
CS Department of Chemistry and Biochemistry, University of Texas, Austin 78712, USA.
NC GM 30048 (NIGMS)
SO BIOCHEMICAL AND BIOPHYSICAL RESEARCH COMMUNICATIONS, (1998 Aug 28) 249 (3)
637-42.

Journal code: 9Y8; 0372516. ISSN: 0006-291X.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 199810
ED Entered STN: 19981020
Last Updated on STN: 19981020
Entered Medline: 19981002
AB Ricin is a heterodimeric cytotoxin composed of RTB, a galactose binding lectin, and RTA, an enzymatic N-glycosidase. The toxin is endocytosed, and after intracellular routing, RTA is translocated to the cytoplasm where it inactivates ribosomes resulting in a loss of host cell protein synthesis and cell death. We show for the first time that the cytotoxicity against cultured T cells by several RTA mutants is directly proportional to the enzyme activity of RTA, suggesting this is a reliable system to measure translocation effects. Large discrepancies between cytotoxicity and enzyme action for a given pair of toxins are therefore attributable to differences in cell binding, uptake, or membrane translocation. Fluid phase uptake and cytotoxicity of isolated RTA are essentially identical to that of the single chain toxin PAP. This important finding suggests that RTA, and the A chain of class 2 RIPs in general, has not evolved special translocation signals to complement the increased target cell binding facilitated by RTB. Experiments with the lectin RCA and with ebulin suggest those toxins have diminished cytotoxicity probably mediated by comparative deficiencies in B chain binding. Addition of a ***KDEL*** sequence to RTA increases fluid phase uptake, consistent with the notion that transport to the ER is important for cytotoxicity. ***Fusion*** of MBP or GST to the amino terminus of RTA has little effect on enzyme action or cytotoxicity. This result is not altered by protease inhibitors, suggesting the ***fusion*** proteins are probably not cleaved prior to translocation of the toxic A chain and implying that the toxins can carry large passenger proteins into the cytoplasm, an observation with interesting potential for analytical and therapeutic chemistry.

L8 ANSWER 41 OF 113 MEDLINE DUPLICATE 23
AN 1998143337 MEDLINE
DN 98143337 PubMed ID: 9484808
TI Design, characterization and anti-tumour cytotoxicity of a panel of recombinant, mammalian ribonuclease-based immunotoxins.
AU Deonarain M P; Epenetos A A
CS Imperial Cancer Research Fund Oncology Unit, Imperial College School of Medicine at the Hammersmith Hospital, London, UK.
SO BRITISH JOURNAL OF CANCER, (1998 Feb) 77 (4) 537-46.
Journal code: AV4; 0370635. ISSN: 0007-0920.
CY SCOTLAND: United Kingdom
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 199803
ED Entered STN: 19980312
Last Updated on STN: 19980312
Entered Medline: 19980305
AB Bovine seminal ribonuclease (BSRNase) is an unusual member of the ribonuclease superfamily, because of its remarkable anti-tumour and immunosuppressive properties. We describe here the construction, expression, purification and characterization of a panel of six immunotoxins based upon this enzyme and show that we can increase its anti-tumour activity by over 2 x 10(4)-fold. This is achieved by improving tumour cell targeting using a single-chain Fv (scFv) directed against the oncofetal antigen placental alkaline phosphatase. As well as the simple scFv-BSRNase ***fusion*** protein, we have constructed five other derivatives with additional peptides designed to improve folding and intracellular trafficking and delivery. We find that the molecule most cytotoxic to antigen (PLAP)-positive cells in vitro is one that contains a C-terminal ' ***KDEL*** ' endoplasmic reticulum retention signal and a peptide sequence derived from diphtheria toxin. All these molecules are produced in Escherichia coli (E. coli) as insoluble inclusion bodies and require extensive in vitro processing to recover antigen binding and ribonuclease activity. Despite incomplete ribonuclease activity and quaternary assembly, these molecules are promising reagents for specific chemotherapy of cancer and are potentially less harmful and immunogenic than current immunotoxins.

L8 ANSWER 42 OF 113 MEDLINE DUPLICATE 24
AN 1998422749 MEDLINE
DN 98422749 PubMed ID: 9750355
TI Stacks on tracks: the plant Golgi apparatus traffics on an actin/ER network.
AU Boevink P; Oparka K; Santa Cruz S; Martin B; Betteridge A; Hawes C
CS Unit of Cell Biology, Scottish Crop Research Institute, Invergowrie, Dundee, UK.
SO PLANT JOURNAL, (1998 Aug) 15 (3) 441-7.
Journal code: BRU; 9207397. ISSN: 0960-7412.
CY ENGLAND: United Kingdom
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 199810
ED Entered STN: 19990106
Last Updated on STN: 19990106
Entered Medline: 19981023
AB We have visualized the relationship between the endoplasmic reticulum (ER) and Golgi in leaf cells of Nicotiana clevelandii by expression of two Golgi proteins fused to green fluorescent protein (GFP). A ***fusion*** of the transmembrane domain (signal anchor sequence) of a rat sialyl transferase to GFP was targeted to the Golgi stacks. A second construct that expressed the Arabidopsis H/ ***KDEL*** receptor homologue aERD2, fused to GFP, was targeted to both the Golgi apparatus and ER, allowing the relationship between these two organelles to be studied in living cells for the first time. The Golgi stacks were shown to move rapidly and extensively along the polygonal cortical ER network of leaf epidermal cells, without departing from the ER tubules. Co-localization of F-actin in the GFP-expressing cells revealed an underlying actin cytoskeleton that matched precisely the architecture of the ER network, while treatment of cells with the inhibitors cytochalasin D and N-ethylmaleimide revealed the dependency of Golgi movement on actin cables. These observations suggest that the leaf Golgi complex functions as a motile system of actin-directed stacks whose function is to pick up products from a relatively stationary ER system. Also, we demonstrate for the first time in vivo brefeldin A-induced retrograde transport of Golgi membrane protein to the ER.

L8 ANSWER 43 OF 113 CAPLUS COPYRIGHT 2001 ACS
AN 1999:36136 CAPLUS
DN 130:219334
TI Differential activity of cholera toxin and E. coli enterotoxin: construction and purification of mutant and ***hybrid*** derivatives
AU Rodighiero, C.; Aman, A. T.; Lencer, W. I.; Hirst, T. R.
CS Department of Pathology and Microbiology, School of Medical Sciences, University of Bristol, Bristol, BS8 1TD, UK
SO Biochem. Soc. Trans. (1998), 26(4), S364
CODEN: BCSTB5; ISSN: 0300-5127
PB Portland Press Ltd.
DT Journal
LA English
AB To det. whether the differential toxicity of cholera toxin (Ctx) and Escherichia enterotoxin (Etx) lies within the A- or B- subunits of the mols., chimeras have been engineered which comprise portions of the A-subunit of Ctx complexed with the B-subunit of Etx and vice versa. A mutant cholera toxin in which the C-terminal ER retention signal (***KDEL***) was substituted for RDEL found in Etx, was also prepd. Here the authors describe the genetic construction of mutant and ***hybrid*** toxins and a method for their purifn.
RE.CNT 6
RE
(1) Amin, T; Prot Expr Purif 1994, V5, P198 CAPLUS
(2) Hirst, T; Handbook of Natural Toxins 1995, V8, P123 CAPLUS
(3) Kaper, J; Nature 1984, V308, P655 CAPLUS
(4) Lencer, W; J Cell Biol 1995, V131, P951 CAPLUS
(5) Mekalanos, J; Meth Enzym 1988, V165, P169 CAPLUS
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L8 ANSWER 44 OF 113 CAPLUS COPYRIGHT 2001 ACS
AN 1998:588578 CAPLUS
DN 129:300572
TI Localization of endoplasmic reticulum in living cells using green fluorescent protein chimeras
AU Van Goethem, Iris D. A.; Adams, Phil; Chad, John E.; Mather, Andrea M.; Griffiths, Barbara; Lee, Anthony G.; East, J. Malcolm
CS School of Biological Sciences, Department of Biochemistry and Molecular Biology, University of Southampton, Southampton, SO167PX, UK
SO Biochem. Soc. Trans. (1998), 26(3), S298
CODEN: BCSTB5; ISSN: 0300-5127
PB Portland Press Ltd.
DT Journal
LA English
AB In order to examine the location of sarcoplasmic/endoplasmic calcium pumps (SERCAs) in COS 7 cells a chimera of SERCA1a and green fluorescent protein (GFP) of Aequorea victoria was produced. In order to det. the location of endoplasmic reticulum (ER) a construct contg. the ER targeting sequence from .alpha.1-antitrypsin attached to GFP terminating with the ER retrieval sequence (***KDEL***) (designated GAP-K) was produced. In order to be certain that the SERCA1a-GFP ***fusion*** protein is correctly targeted the calcium transport properties of the chimera were characterized. SERCA1a-GFP and GAP-K occupied similar internal membrane compartments, presumably ER. A comparison of SERCA1a and SERCA1a-GFP localization indicated that the addn. of GFP to the C-terminus of SERCA1a had not altered its cellular location. The finding that SERCA1a-GFP is able to pump calcium make it unlikely that the ER location of the ***fusion*** protein is the result of mis-folding.

L8 ANSWER 45 OF 113 MEDLINE
AN 1998352821 MEDLINE
DN 98352821 PubMed ID: 9690511
TI BPV-4 E8 transforms NIH3T3 cells, up-regulates cyclin A and cyclin A-associated kinase activity and de-regulates expression of the cdk inhibitor p27Kip1.
AU O'Brien V; Campo M S
CS Beatson Institute for Cancer Research, CRC Beatson Laboratories, Glasgow, Scotland.
SO ONCOGENE, (1998 Jul 23) 17 (3) 293-301.
Journal code: ONC; 8711562. ISSN: 0950-9232.
CY ENGLAND: United Kingdom
DT Journal; Article; (JOURNAL ARTICLE)
LA English

FS Priority Journals
EM 199808
ED Entered STN: 19980828
Last Updated on STN: 19980828
Entered Medline: 19980814

AB The E8 open reading frame of Bovine papillomavirus type 4 (BPV-4) encodes a small (42 amino acid) hydrophobic polypeptide localized to cellular membranes and capable of conferring an anchorage-independent (AI) growth phenotype on primary bovine cells co-transfected with BPV-4 E7 ORF and an activated ras gene. To further study the function of E8 independently of other viral gene products, we have expressed it in the murine fibroblast cell line, NIH3T3. Cells expressing E8 are capable of AI growth and escape growth arrest after serum withdrawal. E8 deregulates cyclin A expression, induces transactivation of the human cyclin A gene promoter and increases endogenous protein levels in cells maintained in short-term suspension culture and in low-serum (LS). Both these culture conditions promote downregulation of cyclin A in control cells. In LS growth conditions E8 permits sustained cyclin A-associated kinase activity but not cyclin E-cdk2 activity. Cyclin A-cdk2 activity and, in part, cyclin A gene expression are regulated by the cdk inhibitor p27Kip1. Expression of this cdk inhibitor is also de-regulated in E8 cells, with high levels being detected under all culture conditions tested. These data suggest that the ability of BPV-4 E8 to transform NIH3T3 cells is associated with upregulation of cyclin A-associated kinase activity and de-regulated expression of the cdk inhibitor p27Kip1 and does not rely on down-regulation of p27Kip1 expression. Analysis of E8 mutants indicate that the hydrophilic 'tail' of the molecule (residues 31-42) is required for cell transformation, as assessed by anchorage-independent growth, while a form of E8 with expression restricted to the Endoplasmic Reticulum/cis-Golgi membranes by addition of a ' ***KDEL*** ' retention signal revealed that the sub-cellular localization is an important determinant of E8 biological activity.

L8 ANSWER 46 OF 113 MEDLINE DUPLICATE 25
AN 1998145457 MEDLINE
DN 98145457 PubMed ID: 9484463
TI Cloning and expression of two genes encoding auxin-binding proteins from tobacco.
AU Watanabe S; Shimomura S
CS National Institute of Agrobiological Resources, Ibaraki, Japan.
SO PLANT MOLECULAR BIOLOGY, (1998 Jan) 36 (1) 63-74.
Journal code: A6O; 9106343. ISSN: 0167-4412.
CY Netherlands
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
OS GENBANK-X70902; GENBANK-X70903
EM 199803
ED Entered STN: 19980326
Last Updated on STN: 19980326
Entered Medline: 19980319

AB Two genes encoding the auxin-binding protein (ABP1) of tobacco (Nicotiana tabacum L.), both of which possess the characteristics of a luminal protein of the endoplasmic reticulum (ER), were isolated and sequenced. These genes were composed of at least five exons and four introns. The two coding exons showed 95% sequence homology and coded for two precursor proteins of 187 amino acid residues with molecular masses of 21,256 and 21,453 Da. The deduced amino acid sequences were 93% identical and both possessed an amino-terminal signal peptide, a hydrophilic mature protein region with two potential N-glycosylation sites and a carboxyl-terminal sorting signal, ***KDEL*** , for the ER. Restriction mapping of the cDNAs encoding tobacco ABP1, previously purified by amplification of tobacco cDNA libraries by polymerase chain reaction (PCR) using specific primers common to both genes, indicated that both genes were expressed, although one was expressed at a higher level than the other. Genomic Southern blot hybridization showed no other homologous genes except for these two in the tobacco genome. The apparent molecular mass of the mature form of tobacco ABP1 was revealed to be 25 kDa by SDS polyacrylamide gel electrophoresis using affinity-purified anti (tobacco ABP1) antibodies raised against a ***fusion*** protein with maltose-binding protein. Expression of the recombinant ABP1 gene in transgenic tobacco resulted in accumulation of the 25 kDa protein. A single point mutation of an amino acid residue at either of the two potential N-glycosylation sites resulted in a decrease in the apparent molecular mass and produced a 22 kDa protein. Mutations at both sites resulted in the formation of a 19.3 kDa protein, suggesting that tobacco ABP1 is glycosylated at two asparagine residues.

L8 ANSWER 47 OF 113 MEDLINE
AN 1998087586 MEDLINE
DN 98087586 PubMed ID: 9425149
TI Retrograde transport of Golgi-localized proteins to the ER.
AU Cole N B; Ellenberg J; Song J; DiEuliis D; Lippincott-Schwartz J
CS Cell Biology and Metabolism Branch, National Institute of Child Health and Human Development, National Institutes of Health, Bethesda, Maryland 20892, USA.
SO JOURNAL OF CELL BIOLOGY, (1998 Jan 12) 140 (1) 1-15.
Journal code: HMV; 0375356. ISSN: 0021-9525.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 199802
ED Entered STN: 19980224
Last Updated on STN: 19980224
Entered Medline: 19980210

AB The ER is uniquely enriched in chaperones and folding enzymes that facilitate folding and unfolding reactions and ensure that only correctly folded and assembled proteins leave this compartment. Here we address the extent to which proteins that leave the ER and localize to distal sites in the secretory pathway are able to return to the ER folding environment during their lifetime. Retrieval of proteins back to the ER was studied using an assay based on the capacity of the ER to retain misfolded proteins. The lumenal domain of the temperature-sensitive viral glycoprotein VSVGtsO45 was fused to Golgi or plasma membrane targeting domains. At the nonpermissive temperature, newly synthesized ***fusion*** proteins misfolded and were retained in the ER, indicating the VSVGtsO45 ectodomain was sufficient for their retention within the ER. At the permissive temperature, the ***fusion*** proteins were correctly delivered to the Golgi complex or plasma membrane, indicating the lumenal epitope of VSVGtsO45 also did not interfere with proper targeting of these molecules. Strikingly, Golgi-localized ***fusion*** proteins, but not VSVGtsO45 itself, were found to redistribute back to the ER upon a shift to the nonpermissive temperature, where they misfolded and were retained. This occurred over a time period of 15 min-2 h depending on the chimera, and did not require new protein synthesis. Significantly, recycling did not appear to be induced by misfolding of the chimeras within the Golgi complex. This suggested these proteins normally cycle between the Golgi and ER, and while passing through the ER at 40 degrees C become misfolded and retained. The attachment of the thermosensitive VSVGtsO45 lumenal domain to proteins promises to be a useful tool for studying the molecular mechanisms and specificity of retrograde traffic to the ER.

L8 ANSWER 48 OF 113 MEDLINE DUPLICATE 26
AN 1998044220 MEDLINE
DN 98044220 PubMed ID: 9382863
TI The mammalian protein (rbet1) homologous to yeast Bet1p is primarily associated with the pre-Golgi intermediate compartment and is involved in vesicular transport from the endoplasmic reticulum to the Golgi apparatus.
AU Zhang T; Wong S H; Tang B L; Xu Y; Peter F; Subramaniam V N; Hong W
CS Membrane Biology Laboratory, Institute of Molecular and Cell Biology, Singapore 119076, Singapore.
SO JOURNAL OF CELL BIOLOGY, (1997 Dec 1) 139 (5) 1157-68.
Journal code: HMV; 0375356. ISSN: 0021-9525.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
OS GENBANK-AF007551; GENBANK-AF007552
EM 199712
ED Entered STN: 19980116
Last Updated on STN: 19980116
Entered Medline: 19971230

AB Yeast Bet1p participates in vesicular transport from the endoplasmic reticulum to the Golgi apparatus and functions as a soluble N-ethylmaleimide-sensitive factor attachment protein receptor (SNARE) associated with ER-derived vesicles. A mammalian protein (rbet1) homologous to Bet1p was recently identified, and it was concluded that rbet1 is associated with the Golgi apparatus based on the subcellular localization of transiently expressed epitope-tagged rbet1. In the present study using rabbit antibodies raised against the cytoplasmic domain of rbet1, we found that the majority of rbet1 is not associated with the Golgi apparatus as marked by the Golgi mannosidase II in normal rat kidney cells. Rather, rbet1 is predominantly associated with vesicular spotty structures that concentrate in the peri-Golgi region but are also present throughout the cytoplasm. These structures colocalize with the ***KDEL*** receptor and ERGIC-53, which are known to be enriched in the intermediate compartment. When the Golgi apparatus is fragmented by nocodazole treatment, a significant portion of rbet1 is not colocalized with structures marked by Golgi mannosidase II or the ***KDEL*** receptor. Association of rbet1 in cytoplasmic spotty structures is apparently not altered by preincubation of cells at 15 degrees C. However, upon warming up from 15 to 37 degrees C, rbet1 concentrates into the peri-Golgi region. Furthermore, rbet1 colocalizes with vesicular stomatitis virus G-protein en route from the ER to the Golgi. Antibodies against rbet1 inhibit in vitro transport of G-protein from the ER to the Golgi apparatus in a dose-dependent manner. This inhibition can be neutralized by preincubation of antibodies with recombinant rbet1. EGTA is known to inhibit ER-Golgi transport at a stage after vesicle docking but before the actual ***fusion*** event. Antibodies against rbet1 inhibit ER-Golgi transport only when they are added before the EGTA-sensitive stage. These results suggest that rbet1 may be involved in the docking process of ER-derived vesicles with the cis-Golgi membrane.

L8 ANSWER 49 OF 113 MEDLINE
AN 97462914 MEDLINE
DN 97462914 PubMed ID: 9323141
TI Visualization of ER-to-Golgi transport in living cells reveals a sequential mode of action for COPII and COPI.
AU Scales S J; Pepperkok R; Kreis T E
CS Department of Cell Biology, University of Geneva Sciences III, Switzerland.
SO CELL, (1997 Sep 19) 90 (6) 1137-48.
Journal code: CQ4; 0413066. ISSN: 0092-8674.
CY United States